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VOLUME LIV NUMBER 6

THE

BOTANICAL GAZETTE

DECEMBER 1912

THE LIFE HISTORY OF CUTLERIA

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 163

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(WITH FIFTEEN FIGURES AND PLATES XXVI TO XXXV)

Introduction

This paper deals with nuclear conditions in *Cutleria multifida* J. Ag. and *Aglaozonia reptans* Crouan, both of which are found in the Bay of Naples. In common with *Zanardinia*, another member of the Cutleriaceae, *Cutleria* and *Aglaozonia* are characterized by having large motile spores. The vegetative body and method of forming reproductive organs show a combination of characters found in *Ectocarpus*, *Sporochnus*, *Tilopteris*, *Sphacelaria*, *Battersia*, *Zonaria*, *Padina*, and *Laminaria*, genera belonging to different families.

The large motile spores attracted the attention of THURET (**63**), who was the first to do experimental work with *Cutleria multifida*. THURET was followed by many investigators who made cultures of the spores in order to determine whether *Cutleria* and *Aglaozonia* might be two alternating generations of one life cycle, but the results were conflicting.

FALKENBERG (**12**), working with material from Naples, first suggested that *Aglaozonia parvula* is the asexual form of *Cutleria multifida*, and that *Aglaozonia chilosa* is the asexual form of *Cutleria adspersa*. CHURCH (**6**), with material from Plymouth, connected *Aglaozonia reptans* with *Cutleria multifida*, and SAUVAGEAU (**43–51**), with material from Guëthary and Banyuls, proved that

Aglaozonia melanoidea is the asexual form of *Cutleria adspersa*. The conclusions of these authors were drawn chiefly from a comparison of young forms of *Cutleria* collected in nature with young plants obtained from cultures, the emphasis being laid upon outer morphological characters.

It is not necessary to present a detailed historical review of previous work, for many authors, especially SAUVAGEAU (**45**), have given such reviews. Since 1899 no work based upon original investigation has been published except by SAUVAGEAU and the present author. Papers by SAUVAGEAU (**46–51**) confirm his work of 1899, and a paper by the author (**76**) is the preliminary account of the present paper. Before turning to our own investigation, we shall indicate briefly what has been accomplished already, and what still remains to be done.

The first definite account of the discharge and the culture of spores of *Cutleria* was made by THURET (**63**), who observed that the female gametes germinated parthenogenetically. The male plants of *Cutleria* he found to be extremely rare at Saint Vaast-la-Hogue, France, where his cultures were made. This parthenogenetic product, a small plant somewhat resembling *Ectocarpus*, did not live long. According to SAUVAGEAU'S suggestions (**45**) the plant may be designated as form *Thuretiana*. Five years later the brothers CROUAN (**7**), by repeating these observations at Brest, France, came to the same conclusions. DERBÉS and SOLIER (**11**), working on the Mediterranean coast of France, confirmed THURET'S account. Thus these earlier observers, who worked on the French coast, seem to have entertained no doubt of the constancy of the parthenogenetic germination of the female gametes of *Cutleria*.

About two decades later two German botanists, REINKE and FALKENBERG, independently making cultures at the Naples Station, reported quite different results. REINKE (**38, 39**), working at the Station in 1875–1876, observed the actual fertilization of the female gametes by the male gametes of both *Zanardinia* and *Cutleria*. In vessels containing both male and female gametes germination took place freely. There seemed to be no germination of the unfertilized egg. REINKE concluded that sexual reproduction prevailed in *Cutleria multifida* and that THURET must have seen only exceptional cases of parthenogenesis. He states that in the Bay of Naples the male and female plants occur in the ratio of 3 to 2. He obtained filaments of both *Zanardinia* and *Aglaozonia* which produced non-motile spores. In the cultures of *Cutleria*, *Desmotrichum* finally appeared, and consequently the ultimate products of the fertilized female gametes were not clearly followed.

FALKENBERG (**12**) in 1878, taking great care to obtain pure cultures of both fertilized and unfertilized female gametes, fully demonstrated the necessity of fertilization for germination. The female gametes, entirely separated from the male gametes, retained their capacity for fertilization for four or five days and then never grew beyond the formation of a thin cell membrane. FALKENBERG not only demonstrated the necessity for fertilization, but succeeded in developing the sporelings to a considerable size; in 6–8 weeks the sporeling in the stage of "foot embryo" (*Keimfuss*) had developed as a secondary lateral outgrowth the creeping flat form (*kriechende Flachsprosse*), which is identical with the creeping thallus of *Aglaozonia*. This kind of sporeling may be designated as form *Falkenbergiana*. FALKENBERG was the first to associate *Cutleria multifida* with *Aglaozonia reptans*, and *Cutleria adspersa* with *Aglaozonia chilosa*. Four years later JANCZEWSKI (**21**), at Antibes, did with *Cutleria adspersa* what REINKE and FALKENBERG had done with *Cutleria* at Naples, and showed that unfertilized gametes of *Cutleria adspersa* do not develop further.

In 1894 KUCKUCK (**24**) described a plant under the name of *C. multifida* var. *confervoides*. The plants came up in the tank of the Helgoland Laboratory in the summer of 1893, and were attached to stones which had been collected in the North Haven in fairly shallow water of 1–3 fathoms. The plants were monosiphonic and some of them were in the reproduction stage. The normal *Cutleria* is said to have been gathered at Helgoland by WOLLNY, but has not been known to occur there since. The origin of the plant is not clear.

The cultures of *Cutleria* and *Aglaozonia* made by CHURCH in 1898 were from material collected in English waters. CHURCH (**6**) obained the material by dredging in the estuary of the river Yealm, near Plymouth, England, at 2–3 fathoms below low water mark. The male plants of *Cutleria multifida* were very rare, and in cultures of the female gametes there was produced parthenogenetically a form identical with the form *Falkenbergiana*. Further, from the zoospores of *Aglaozonia parvula* CHURCH obtained plants which, like the preceding, have the creeping platelike thallus of *Aglaozonia*, but whose column ends at the summit in filaments (not fascicled) which bear the male gametangia of *Cutleria*. The latter, being entirely a new form and having never been obtained by any previous investigators, may be designated as form *Churchiana*. Finding that the female gametes develop parthenogenetically and the zoospores of *Aglaozonia* produce *Aglaozonia* directly with no alternating appearance of *Cutleria*, CHURCH concludes that *Cutleria* and *Aglaozonia* represent simply polymorphism due to environmental conditions, especially such as changes in the temperature of water.

In 1898 SAUVAGEAU (**43**) reported that he found in nature, epiphytic upon adult *Cutleria adspersa*, a number of young sporelings of the same species. These sporelings were of two kinds, forms *Thuretiana* and *Falkenbergiana*, which were evidently growing side by side, at the same time, and on the same spot under similar conditions. In the summer of 1898, KUCKUCK (**25**) obtained in cultures at the Helgoland Laboratory a number of young plants germinating from the zoospores of *Aglaozonia parvula*. The young plants had the characters of the form *Thuretiana*, and in his material female gametangia of *Cutleria* were produced. Besides, some of the plants produced a creeping disk, so that the whole structure resembled the form *Churchiana*, but the form *Falkenbergiana* never appeared.

SAUVAGEAU (**44, 45**) found at several points on the coast of the Gulf of Gascony, France, a new *Aglaozonia*, the *Zonaria melanoidea* Schousboe, discovered at the beginning of the century at Maroc and not reported since, whose *Aglaozonia* nature remained unrecognized. SAUVAGEAU considers *Aglaozonia melanoidea* to be the sporophyte of *Cutleria adspersa*. In cultures of *Cutleria adspersa* which he collected at Guéthary (Basses-Pyrénées), he observed that the female gametes did not attract the male gametes. The female gametes germinated very readily parthenogenetically and gave always and characteristically the form *Falkenbergiana*. In 1905 SAUVAGEAU (47, 48) collected *Aglaozonia melanoidea* at Banyuls-sur-Mer (Pyrénées-Orientales), and in winter and early spring in each successive year of 1905–1908, he (46–51) carried on his cultures at the Laboratory Arago, Banyuls. From the zoospores of *Aglaozonia melanoidea* he obtained, in a majority of cases, *Cutleria adspersa*, and in only one case out of 100 did he obtain *Aglaozonia melanoidea;* but neither the form *Churchiana* nor the form *Kuckuckiana* was produced. Unfertilized female gametes of *Cutleria adspersa* produced either *Aglaozonia* or *Cutleria*.

From his own results and those of previous investigations, SAUVAGEAU concluded that the alternation of two generations is not necessary, but rather, as it might be said, facultative. OLTMANNS (35), from results of previous authors, considers *Cutleria* as presenting no true alternation with the *Aglaozonia* generation, but as simply presenting another instance of polymorphism in algae. STRASBURGER (59), discussing the alternation of generations in Phaeophyceae, agrees with the views of SAUVAGEAU and OLTMANNS.

All these investigations, discussions, and conclusions were made without any reference to nuclear conditions. The cytology of *Cutleria* and *Aglaozonia* was first described by the author as a preliminary note three years ago (74).

The material for the present investigation was collected at the Bay of Naples in the winter of 1908 and spring of 1909, during which time I occupied a table of the Carnegie Institution at the Stazione Zoologica. *Cutleria multifida* was found growing on rocks at a depth of 1–5 meters in the vicinity of Posilipo and Castello dell 'Uovo, and *Aglaozonia reptans* was growing on the surface of echinoderm shells or stones in the same localities and also at Nisida along the Bay of Naples, often at a greater depth. A fresh and plentiful supply of material was furnished by Dr. SALVATORE LO BIANCO almost every day. Cultures of the plants and of their sporelings and the fixation of their critical stages were made in the laboratory of the Station, where by the kindness of Drs. ANTON DOHRN and REINHART DOHRN the author enjoyed every equipment for facilitating the work. The investigation thus begun at Naples was finished at the Hull Botanical Laboratory of the University of

Chicago. To Professor JOHN M. COULTER and Professor CHARLES J. CHAMBERLAIN I wish to express my thanks for their suggestions and criticisms throughout the investigation.

The paper presents the results of my studies on *Cutleria* and *Aglaozonia*. For *Cutleria* there is described the mitosis in the vegetative cells of the male and female plants, the formation of the male and female gametes, the fertilization and the germination of the fertilized female gametes, and the germination of the unfertilized female gametes; for *Aglaozonia* the mitosis in the vegetative cells, and the formation and germination of the zoospores. Finally, there is a brief discussion of the cytological phenomena and the alternation of generations.

Cutleria multifida

Cutleria multifida J. Ag. is generally dioecious, and the forms of the male and female plants vary with the localities in which they grow. At one place, the male plants are more broadly multifid and shorter than the female, which are very narrowly multifid and often reach a length of 25 cm. or more; in other localities the reverse is true, that is, the male plants are narrowly divided and larger than the female plants. An extensive comparative study of the forms shows that there is great variability in habit, so that it seems impossible to distinguish the two mature sexual individuals by any gross morphological aspects, except that they bear, as a rule, exclusively either male or female reproductive organs. *Cutleria* grows in the Bay of Naples upon rocks or mollusk shells, at a depth of 1–5 meters. The *Cutleria* material was collected with the rocks or shells upon which it was growing, and there was always an abundant display of the successive stages in the development of the plants, from the very young thallus to large adult forms. The young thallus is 1–3 mm. in length, narrowly funnel-shaped, and presents no feature to distinguish the male plants from the female.

MITOSIS IN THE VEGETATIVE CELLS OF THE MALE AND FEMALE PLANTS

Both male and female plants, when fresh, always bear a beautiful fascicle of hairs at the tips of the multifid filaments of the thallus, and scattered here and there upon the whole surface of the thallus there are also tufts of hairs. Any part of the frond in vigorous growth is favorable for the study of vegetative mitosis, but details are more easily and definitely followed in the hairs and in the superficial layer of the thallus.

The cells in these regions are full of plastids, with usually a single nucleus in the center. The nucleus in the resting stage is very small, generally about the size of the plastids or slightly larger (figs. 1, 5, 16, 53). The network is so fine that it is hard to recognize much chromatin in it. Neither centrosome nor central bodies with or without radiations seem to be present.

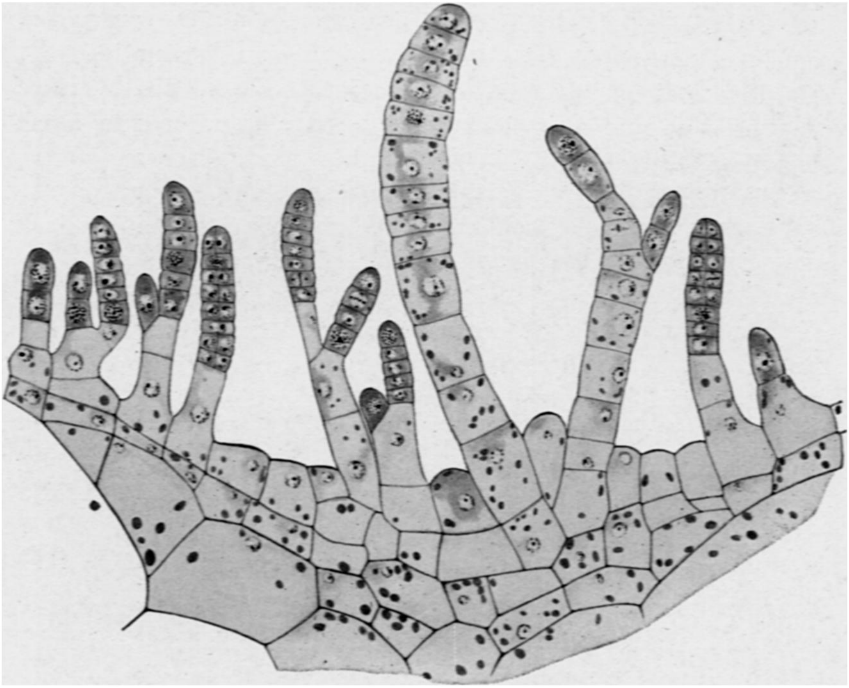


FIG. 1.—*Cutleria multifida:* portion of a thallus with a number of filaments and young male gametangia.

In early prophase the nucleus increases in size, until it becomes three or four times the diameter of the resting nucleus and is a conspicuous structure within the cell. During the growth of the nucleus, just inside the membrane, there appear chromatin knots which are evidently worked out of the chromatin network by the rearrangement of the material. These chromatin knots, which are continuous with less deeply stained chromatin fibrils, are variable in number at first, but gradually the number of chromatin knots becomes definite and they are detached entirely from the chromatin fibrils and become the chromosomes, 24 in number (figs. 19, 54). The chromosomes, after segmentation, gradually assume a slightly elongated rod form and become arranged at the equatorial plate (figs. 20, 55).

A little before the equatorial plate stage, from the cytoplasm surrounding the nuclear membrane, kinoplasmic accumulations arise at the poles. A well marked centrosome-like structure in the kinoplasmic masses occurs only at the metaphase. The chromosomes split longitudinally and half of each chromosome proceeds to each pole (figs. 22, 56). During this entire process the spindle is intranuclear. At telophase the nuclear membrane disappears and the two sets of daughter chromosomes, crowded closely together, are surrounded by cytoplasm, and the formation of the nuclear membrane follows (figs. 28, 57).

When the daughter nuclei are organized, the central spindle disappears completely (figs. 29, 57). The cytoplasm lying between the two nuclei begins gradually to assume a coarse, irregular, alveolar structure, and the walls of the alveoli, probably after a change in their material, form a new cell plate. Vegetative mitosis is essentially the same in both male and female plants.

The size of the nucleus varies according to the portion of the thallus in which the nucleus is contained. The hairs and superficial cells contain larger nuclei than the huge cells situated below the superficial cells; however, even those in the hairs and superficial cells vary in size. There seems to be no relation between the size of the nucleus and that of the cell. Quite frequently the narrow cells in the hairs contain larger nuclei than the elongated cells; usually the marginal cells in the superficial layer of the thallus have larger nuclei than the others. On the whole, the nuclei of the female plants are slightly larger than those of the male plants. However, such a difference, if it exists, is not very great during the vegetative division, but the conspicuous difference in size appears in the formation of the reproductive organs.

THE MALE GAMETANGIUM

Mature male gametangia.—Male gametangia occur all over the surface of the thallus in small or in large clusters (figs. 1, 2, 3, 4). When the male plant is young, before the appearance of gametangia, the surface of the thallus bears tufts of hairs here and there in somewhat regularly scattered spots. Later, with or without the association of the hairs, young male gametangia appear. Both the male gametangia and hairs arise from superficial cells of the thallus.

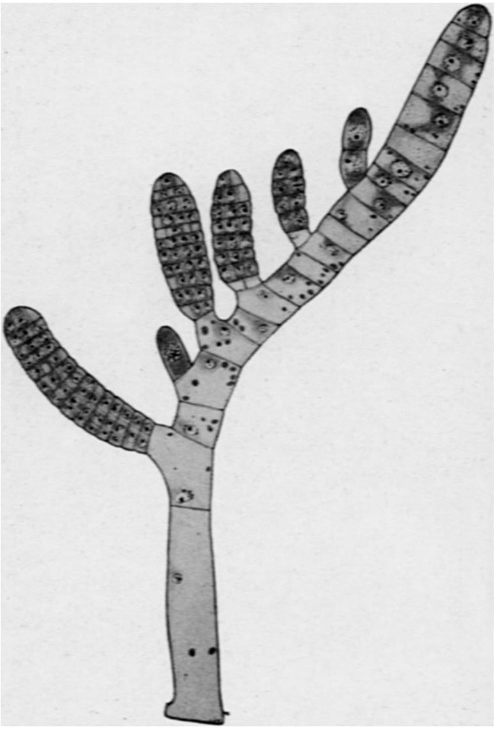
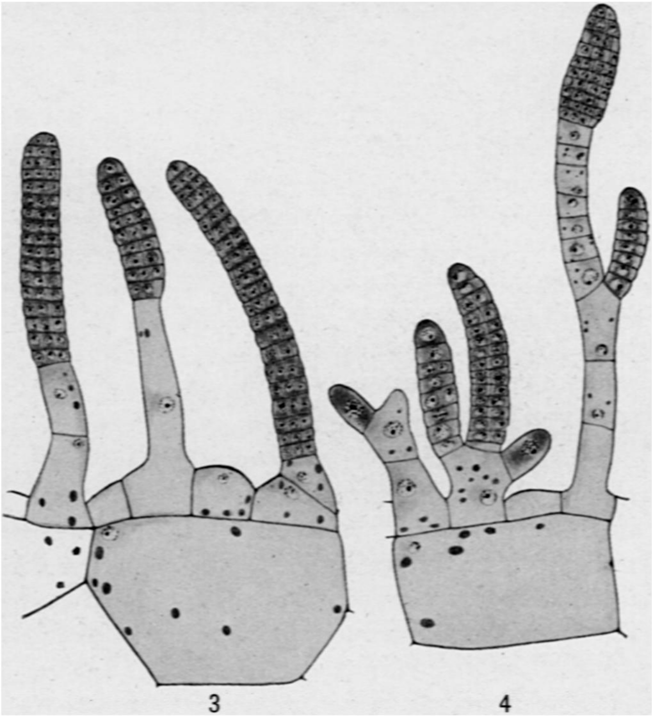


FIG. 2.—*Cutleria multifida:* a filament bearing a few male gametangia in successive stages of development.

The mature male gametangium consists of a number of tiers of small cells (the male gamete mother cells), each tier composed of 8 cells, and since there are more than 22 tiers (fig. 4), the output of male gametes from a single gametangium is about 200. The mature male gamete in the free swimming condition outside of the gametangium has an oval shape (fig. 8, *a*) and usually contains two plastids. Associated with one of the plastids, inside of the lateral face of the plasma membrane near the anterior end, is a red pigment, and in close association with this pigment are two cilia of different lengths, the long cilium directed toward the anterior end, 4.5 times the long diameter of the gamete, and the short one in the opposite direction, about twice the diameter of the gamete. The long diameter of the entire male gamete is 5 μ (figs. 3 and 4).

Development of male gametangia.—As was stated above, both the hairs and gametangia arise from the superficial cells of the thallus.

One of the superficial cells commences to grow more vigorously than the rest (fig. 17) and a typical nuclear division takes place, which gives rise to a gametangium initial and a stalk cell. Sometimes 2–7 or more subsequent divisions occur and there is produced a filament of two or more cells, the terminal one of which is destined to be a male gametangium initial (figs. 1, 3, 4). Or a filament becomes a long multicellular structure of a single row of cells, and from a number of these cells there are produced lateral branches, consisting of one, two, three, or more cells, the terminal one of which becomes the gametangium initial (figs. 2, 21, 39).



FIGS. 3, 4.—*Cutleria multifida:* fig. 3, one of the male gametangia, with a long stalk composed of several cells; fig. 4, gametangia near maturity; one of them has 22 tiers of mother cells.

The nucleus of the male gametangium initial is considerably larger than is common in cases of vegetative mitosis (fig. 18). The chromatin network of the resting nucleus is marked by a number of knots mixed with broken fibrils (fig. 18). The chromatin knots increase gradually in size in prophase and finally break up into 24 chromosomes (fig. 19). Up to this stage the nucleolus remains without apparent change. When the chromosomes begin to be arranged at the equatorial plate, the nuclear membrane becomes contracted, and at the completion of the nuclear figure the decrease of the diameter of the nucleus is remarkable (fig. 20). Two distinct centrosome-like structures now are present at metaphase. While the nuclear membrane still persists, the chromosomes split longitudinally and half of each chromosome proceeds to each pole (fig. 22). The nucleus passes into telophase, two new daughter nuclei are formed, and then the subsequent divisions occur. In the second division the two nuclei may undergo mitotic changes simultaneously, both showing 24 chromosomes in prophase (fig. 23), or one may proceed more rapidly than the other. Figs. 24 and 25 show the latter case; the nucleus of the upper cell is in prophase, showing 24 chromosomes, and that of the lower cell is in metaphase. In figs. 24 and 25, in spite of the apparent similarity in the size of the lower cells, the difference in the size of their nuclei at metaphase is remarkable. Sometimes the nucleus of the upper cell is far behind that of the lower cell in dividing, so that, as in fig. 26, the upper nucleus is in the resting condition while the lower one is in anaphase. Fig. 27 shows the nucleus of the upper cell in metaphase, which passes to anaphase (fig. 28) and to telophase (fig. 29) while that of the lower cell remains resting. When the filament has reached the 3-celled stage mitosis in the three nuclei proceeds in varying order. In fig. 30 is shown the case in which the nucleus of the basal cell is more advanced, revealing 24 chromosomes in prophase. The following figure shows the nucleus of the terminal cell in advance and now in metaphase. In the next figure the nucleus of the middle cell is already in anaphase.

At the 4-celled stage of the young gametangium, the nucleus in prophase invariably shows 24 chromosomes (figs. 33, 34). At about this stage the nucleus in division has almost the same size and aspect as the nuclei in vegetative filaments. In figs. 35 and 36 are given anaphase and prophase of the vegetative division. Until about the 4 or 5-celled stage the young gametangium consists of a single row of cells, after which division also occurs perpendicular (figs. 37, 48) to the direction of the axis of the gametangium, so that there are two rows of cells. When the young gametangium has reached the 8 or more-celled stage and consists of two rows of cells, the nucleus in both the resting and dividing condition has necessarily been forced to assume a somewhat irregular form so as to occupy the largest possible space, and it does not decrease much in size in spite of the multiplied divisions and consequent diminution of the cells of the gametangium (figs. 40, 41).

Another division perpendicular to the direction of the growth of the gametangium occurs (figs. 42, 44), and the gametangium becomes a structure composed of four rows of cells. The diminution in the size of the nucleus is now recognizable (figs. 43, 45). Although the chromosomes which appear in prophase are 24 in number, their size has become remarkably reduced to about half the diameter of those appearing in earlier mitoses. The diminution may proceed still further during the divisions which multiply the number of the cells in a tier. There now occurs the third and last division perpendicular to the axis of growth of the gametangium (fig. 50), resulting in a structure composed of eight rows of cells. Even after the gametangium shows eight rows of cells, a number of mitoses may occur that increase the number of cells in the rows (figs. 46, 47, 48, 49), and during these mitoses there are always 24 chromosomes. These longitudinal divisions of the gametangium, as well as the transverse, are strictly simultaneous, but they proceed at a nearly uniform rate, so that ultimately there is formed the well known male gametangium of *Cutleria*, which is composed of eight rows of cells, each row made up of a varying number of cells. The number of cells in a row may be even more than 24, the cells of each row lying side by side with the cells of the contiguous row. Each individual cell in the gametangium is a male gamete mother cell, within which a single male gamete is formed.

The mother cell contains a single large nucleus situated in the center, and usually two plastids whose position varies. The nucleus passes into a completely resting condition. Fig. 52 represents a cross-section of a male gametangium, each gamete mother cell showing a red pigment spot in close association with the nucleus. When the male gametes are mature within, a portion of the free surface of the membrane of the mother cell dissolves so as to leave a small pore, through which the gametes are discharged. The cilia of the gametes first protrude from the pore, keep moving for a while, and then all the contents emerge and the male gamete is set free.

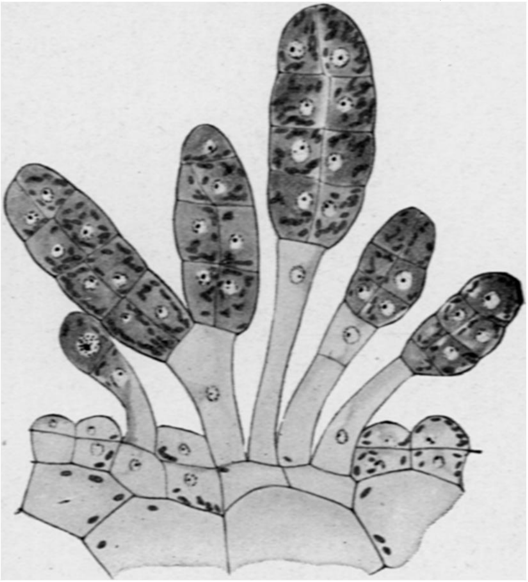
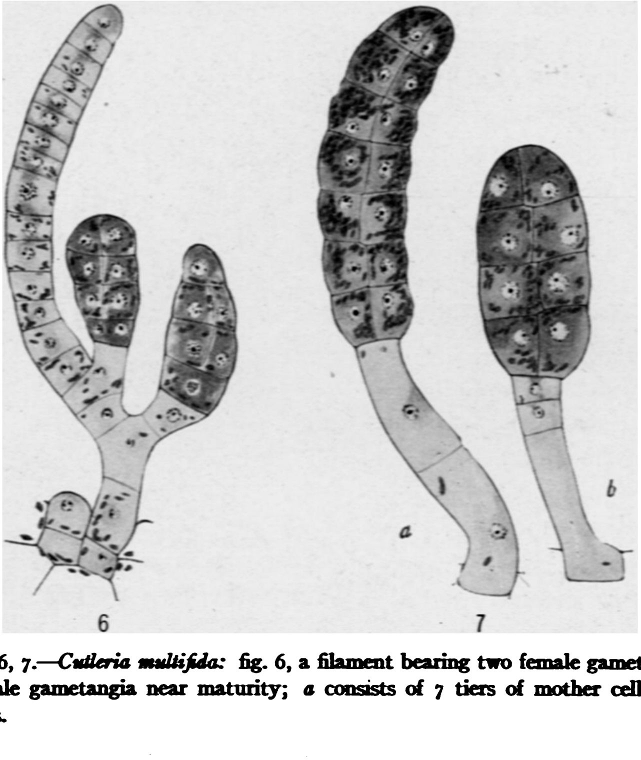


FIG. 5.—*Cutleria multifida:* portion of a thallus with a number of filaments bearing female gametangia in various stages of development.

THE FEMALE GAMETANGIUM

Mature female gametangia.—Female gametangia are scattered over the whole surface of the thallus, as in the case of the male gametangia (figs. 5, 6, 7). The female gametangia are developed alone or are associated with hairs which appear upon the surface of the thallus long before the appearance of the gametangia. As in the case of the male plants, the hair and gametangium originate from superficial cells of the thallus.

The mature female gametangium consists of several tiers of female gamete mother cells, the number of tiers varying from 4 to 7. Each tier is composed of 4 or 8 mother cells, so that the output of gametes from a single female gametangium fluctuates between 16 and 56. The mature female gamete in the free swimming condition outside of the gametangium is oval (fig. 8, *b*), and usually contains more than 30 plastids. The anterior end of the body of the female gamete is destitute of plastids and contains colorless granular cytoplasm, thus indicating the existence of polarity in the distribution of plastids. In association with one of the plastids, which is situated near the periphery in the vicinity of the anterior end, there is present a red pigment spot. Close to the pigment spot, two cilia of unequal length are borne; the long one being directed toward the anterior end, 1.5 times the long diameter of the gamete, and the short one in the opposite direction about equal in length to the diameter of the gamete. The long axis of the female gamete is 26.2 μ. The female gamete is actively motile when discharged, but after a while the movement becomes sluggish, the shape becomes spherical (fig. 8, *c*), and the cilia are withdrawn.



FIGS. 6, 7.—*Cutleria multifida:* fig. 6, a filament bearing two female gametangia; fig. 7, female gametangia near maturity; *a* consists of 7 tiers of mother cells, and *b* of 4 tiers.

Development of female gametangia.—Like the male gametangium, the female gametangium arises from a superficial cell of the thallus. One of the superficial cells begins to grow more rapidly than the others (fig. 53) and a typical mitosis takes place, which gives rise to a gametangium initial and a stalk cell. Sometimes a second or third division is intercalated between the first division occurring in the superficial cell and the formation of the gametangium initial, and in this case the female gametangium, instead of having a stalk of a single cell, has a stalk consisting of two (fig. 7, *a*) or three cells (fig. 7, *b*). Or a number of divisions follow the first, so that the superficial cell develops into a long filament and some cell of the filament divides laterally to form a branch which becomes a gametangium initial (fig. 6).

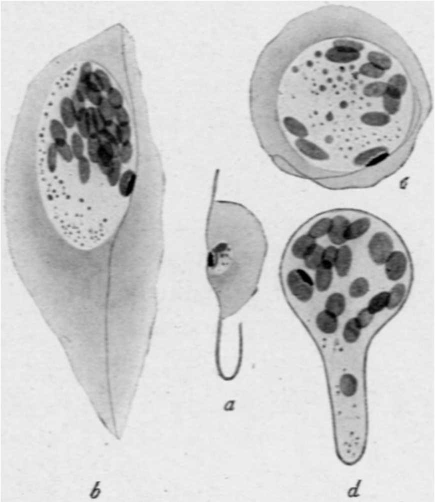


FIG. 8.—*Cutleria multifida:* gametes sketched from living material; *a*, male gamete; *b*, female gamete; *c*, female gamete which has become quiescent and spherical; *d*, female gamete germinating apogamously.

The nucleus of the gametangium initial increases greatly in size, as in the case of the male gametangium initial. In it there is contained a large number of plastids. The chromatin network of the nucleus, which is composed of irregular knots and scanty fibrils during the resting stage, gradually increases in chromatin content, and finally there is established a prophase (fig. 54) containing 24 chromosomes and a single nucleolus. In metaphase the volume of the nucleus diminishes considerably (fig. 55), when compared with the previous stage, and centrosome-like structures appear at the poles. While the nuclear figure is still intranuclear, the chromosomes split longitudinally and half of each chromosome passes to each pole (fig. 56). At telophase two daughter nuclei are formed, and no central spindle remains between them. The cell plate is laid down by the cooperation of the vacuoles and rearrangement of the cytoplasm (fig. 57). Each nucleus of the two cells grows in size simultaneously during the resting condition (fig. 58) and then the rates of the mitotic processes in these two nuclei diverge; the upper nucleus enters prophase, showing 24 chromosomes (fig. 59), metaphase with two centrosome-like structures (fig. 60), anaphase (fig. 61), and reaches telophase (fig. 62) while the lower nucleus still remains in the resting condition. Sometimes the lower nucleus divides first, showing metaphase with two centrosome-like structures (fig. 63) and anaphase (fig. 64) while the upper nucleus rests.

In the young female gametangium, consisting of three cells, the mitoses occur in varying order; sometimes the nucleus of the basal cell divides first (figs. 65, 66), sometimes that of the upper cell (figs. 67, 68), and sometimes that of the middle cell (figs. 69, 70), and during these divisions 24 chromosomes are always seen in prophases (figs. 65, 66, 69, 71).

Up to the 4-celled stage the female gametangium divides transversely, and then a perpendicular division occurs (fig. 72), so that the gametangium consists of two rows of cells, after which another longitudinal division (figs. 73, 78) immediately follows and results in producing a gametangium of four rows of cells.

Mitosis in the gametangium of the 7–11-celled stage is not simultaneous, but occurs at different times in different cells. The nucleus in the middle cell may divide ahead of the others (figs. 74–77, 79, 80, 83, 84), whose nuclei are either in the resting stage or in prophase. The nucleus in prophase in any of these cells always presents 24 chromosomes, and in metaphase two centrosome-like structures are seen at the poles. It is remarkable that the chromatin knots or globules in the resting nucleus always lie quite independent of one another, the chromatin fibrils being so scanty that they are unable to connect them together.

In the young gametangia of the 10–18-celled stage, the mitoses take place as in earlier stages (figs. 85–88); the prophase and the polar view of metaphase show clearly that the number of chromosomes is 24. One striking feature as contrasted with the male gametangia is that in the female gametangia the nuclear diminution in size is not very great during the repeated divisions and consequent multiplication of cells of the gametangia. Of course the nucleus at the time of the first division in the female gametangium is rather large, especially in prophase (fig. 54), and the nucleus in the prophase of the division in the female gametangium of the 18-

celled stage is almost two-thirds of that diameter (fig. 87), but if the nuclei in metaphase and in the resting condition are taken into consideration, almost all of the cases in the gametangia from the 2-celled stage to 18-celled stage seem alike. The female gametangium, consisting of four rows of cells, becomes mature (fig. 89), but quite often each of the cells in the four rows divides again, so that the gametangium consists of eight rows of cells (fig. 90).

These numerous nuclear divisions are not simultaneous, but follow in regular order, both those which are transverse to the axis of the gametangium and those which are perpendicular to it, so that the ultimate result is the well known female gametangium of *Cutleria*, which is composed of four or eight rows of cells, each row composed of 4–7 cells, the cells of each row lying almost exactly opposite the cells of the neighboring row. Each individual cell of the gametangium is a female gamete mother cell. The mother cell contains a large nucleus either in the center or near the inner wall (figs. 90, 92). Plastids are crowded either near the inner wall (fig. 91) or near the outer side (fig. 92), or lie scattered regularly throughout the whole cytoplasm. The nucleus is in the resting condition.

When the gametangium is mature, the whole contents of a gamete mother cell become a single female gamete. A portion of the free surface of the mother cell dissolves and forms a pore through which the gamete is discharged. The slow process of discharging the gametes is the same as in the case of the male gametes; first, the cilia of the gametes appear outside the pore, keep waving for a while, and then the female gamete is set free.

FERTILIZATION AND THE GERMINATION OF THE FERTILIZED FEMALE GAMETES

The discharge of both male and female gametes occurs at almost any time during the day and night. However, so far as the writer's experience goes, while he was making observations at half hour intervals under the microscope, the majority of cases showed that the discharge was most abundant at 5 A.M., and that it continues, though gradually diminishing, until 7 A.M. and then ceases. Occasionally there is some discharge at 11 A.M. and 5 P.M. The motile power of the male gametes continues for almost 20 hours, while that of the female gametes is comparatively short, the longest period of the swimming condition observed having been 2 hours, and the shortest 5 minutes. Toward the end of the motile condition in both male and female gametes, the movements become sluggish, and then cilia are no longer recognizable, seemingly being withdrawn or coalescing with part of the plasma membrane of the gamete body, and finally the shape of the body becomes spherical. However, in a number of cases gametes caught in the thallus of algae while swimming between the slide and cover-glass remained active for more than 24 hours, and then became spherical; when stained at that time they showed the cilia still persisting and not coalescent with the plasma membrane.

The union of male and female gametes and the subsequent nuclear behavior were studied in material from artificial cultures. A number of dishes were prepared containing either male or female plants bearing mature gametangia. When the discharge of gametes became abundant, the sea water of the dish containing thousands of swarming male gametes was added to a dish containing vigorous female gametes. Then part of this mixture was observed under the microscope in living condition and part was fixed every 30 minutes for 24 hours, and then at 26, 28, 32, and 48 hours, and later, every 3 or 5 days up to 75 days. The following description is based upon material obtained in this way.

The male gametes while swimming freely become attached to the female gametes which are either sluggishly swimming or have come to rest, but have not yet formed a cell wall. When the male gamete becomes quiescent, before coming in contact with the female gamete it withdraws its cilia and its shape becomes spherical. The nucleus is full of chromatin which is clearly broken into 24 independent chromosomes. The nuclear membrane is very delicate (fig. 94). When a male gamete has just become attached to the female gamete, both gametes have very delicate plasma membranes. The nucleus of the female gamete is at the center of the cell, as in the resting condition (fig. 95).

Then the plasma membranes which lie between the cytoplasm of the male and female gametes become obscure and the cytoplasm of the two gametes comes into direct contact. The male gamete can be observed for a short time as a protuberance at the periphery of the female gamete (fig. 95); however, sooner or later the protuberance is entirely absorbed or leveled down to the surface of the now spherical female gamete. The male nucleus with 24 distinct chromosomes proceeds toward the resting female nucleus (fig. 96); it then proceeds still further (fig. 97) until it is close to the female nucleus (figs. 98, 99). The male nucleus at this stage is surrounded by a clear zone, which possibly means that the nucleus is carrying with it a part of the male cytoplasm or a part of the female cytoplasm which had been lying in the path of the male nucleus. The male nucleus now becomes closely applied to the female nucleus (fig. 100). The chromatin of the male nucleus becomes closely aggregated (fig. 101), and then begins to be finely alveolated (fig. 102), and finally becomes dispersed through the female nucleus as irregular knots (fig. 103). Later the fusion nucleus contains chromatin knots of various sizes and shapes, together with delicate, irregular, discontinuous fibrils. A nucleolus which was present in the female nucleus before the union still persists (fig. 104). The fusion nucleus thus passes into a completely resting condition, in which chromatin of male and female origin cannot be distinguished from each other.

Returning to the previous stage, at the time of the union both gametes are surrounded by plasma membranes only. At the time of the entrance of the male nucleus into the female cytoplasm the outermost layer of the plasma membrane is observed to change into cell wall (fig. 96), which is very thin at first, but gradually increases in thickness as fusion progresses, and is well organized by the time the fusion nucleus reaches the resting condition.

The first segmentation division of the sporelings of the fertilized gametes takes place 20–24 hours after the union of the gametes. In prophase there appear 48 chromosomes, all of which seem alike both in size and shape (fig. 106), and a single nucleolus persists. At metaphase the chromosomes are arranged at the equatorial plate and two centrosome-like structures appear at the poles (fig. 107). A comparison of the prophase and metaphase shows that the volume of the nucleus is considerably diminished at the latter stage. The mitosis may take place before the sporeling begins to elongate; but in the majority of cases the sporeling elongates into a somewhat pear-shaped structure while its nucleus is still in the resting state (fig. 108). Quite often the nucleus is present at the elongated portion (fig. 109). The cell wall of the elongated portion which is to become the holdfast is comparatively thick.

The number of chromosomes at prophase (figs. 110, 111) and seen in polar view at metaphase (fig. 114) is 48. The axis of the figure is often perpendicular to the growing axis of the sporeling (fig. 113). After telophase the sporeling is divided into two cells (fig. 115). The nuclei of the two cells divide one after the other; sometimes the lower nucleus increases more rapidly than the upper (fig. 117), or the upper nucleus begins to divide first (figs. 116, 118–120). In sporelings consisting of three cells, mitosis occurs in various order in the three nuclei; the nucleus of the basal cell may divide first (figs. 121, 122), or the three nuclei may divide simultaneously (fig. 123). The number of chromosomes appearing at the prophase (figs. 121, 123) is 48.

The fourth, fifth, and subsequent divisions, that occur continuously for more than ten days, multiply the number of the cells of the sporeling, resulting in the development of the upright columnar form that grows standing upon the substratum, and then the direction of growth becomes changed.

Before describing this change of growth which occurs in sporelings about ten days old, we shall note briefly the fate of the red pigment that is conspicuous in the living gametes. As was stated before, the red pigment is always associated with the plastid. A part of the plastid becomes impregnated with some substance of a deep orange color, which seems to disorganize instantly with the death of the sporeling. The most interesting point is that the cilia are attached to the pigment, and the red pigment is the only structure within the sporeling that bears any close and direct physical connection with the cilia. Moreover, the plastid that bears the red pigment always lies near the nucleus. By the union of male and female gametes the red pigment bearing plastid of the male gamete is introduced into the female gamete and consequently two red pigment structures are always present in sporelings resulting from fertilization: a large red pigment spot is associated with a large plastid of the female gamete, and a small red pigment spot upon a small plastid is characteristic of the male gamete. Although the plastid bearing the red pigment of the male gamete grows in size after its introduction into the cytoplasm of the female gamete, yet it seldom becomes equal in size. This difference in size of the two pigment spots is maintained as far as the present observation goes. Fig. 9 represents some of the stages in the development of sporelings from the 1-celled to the 5-celled stage, indicating the various positions of the two red pigments of male and female origin. The red pigment was recognizable even up to the 16-celled stage, and beyond the 20-celled stage it becomes hard to recognize, probably on account of the disintegration of the coloring matters. It seems probable that the red pigment is concerned with the motility of the gamete rather than with fertilization.

As stated before, the sporelings develop continuously in one direction, and as a consequence after about ten days there is produced a columnar structure standing upright upon the substratum, and then the direction of the growth becomes changed. Cells of the basal portion of the columnar sporeling divide laterally, instead of in the direction of the axis of the growth, so that by repeated cell division there is produced a flat expansion, the whole structure of which might be well compared with a candlestick, the candle being the column and the base the newly developed flat expansion. The basal expansion is developed by the repeated periodic cell divisions on the lateral margin, which causes a zonation like that in *Peyssonnelia;* besides, the thallus is characterized by intercalary growth. Sporelings at about 20 days after fertilization are shown in fig. 10, *a*. Later the upright column does not seem to grow much, only the basal expansion continuing to develop. Sporelings 30 days old are shown in fig. 10, *b*. In 55 days old sporelings, the growth of the column has ceased, and it remains small, hardly recognizable to the naked eye, while the basal expansion reaches a considerably larger size and becomes well fitted to flourish as an independent creeping structure (fig. 10, *c*, *d*).

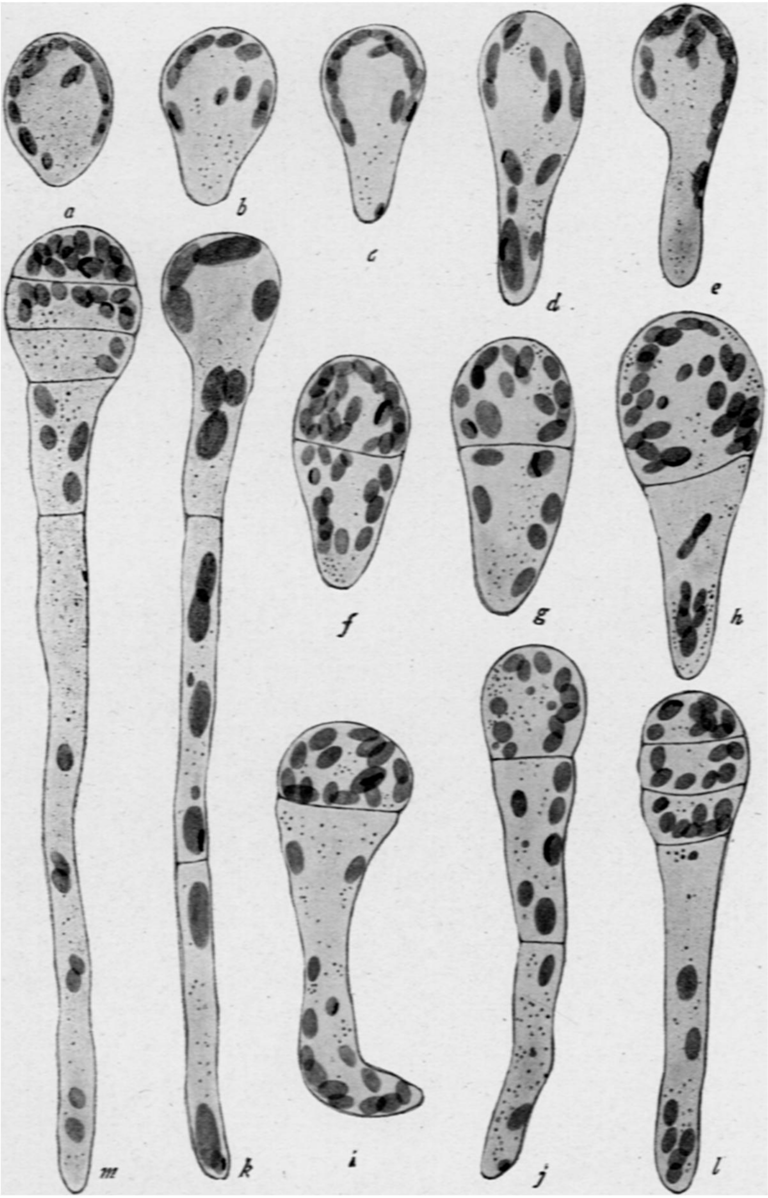


FIG. 9.—*Cutleria multifida:* normal sporelings sketched from living material; *a–e*, one-celled stage; *f–i*, two-celled stage; *j* and *k*, three-celled stage; *l*, four-celled stage; *m*, five-celled stage; nucleus not recognizable in living material; the two pigment spots associated with plastids are of maternal and paternal origin; the small pigment spot introduced by the male gamete, the large one by the female gamete; the plastid that bears the small red pigment spot has increased in size after its introduction into the female gamete.

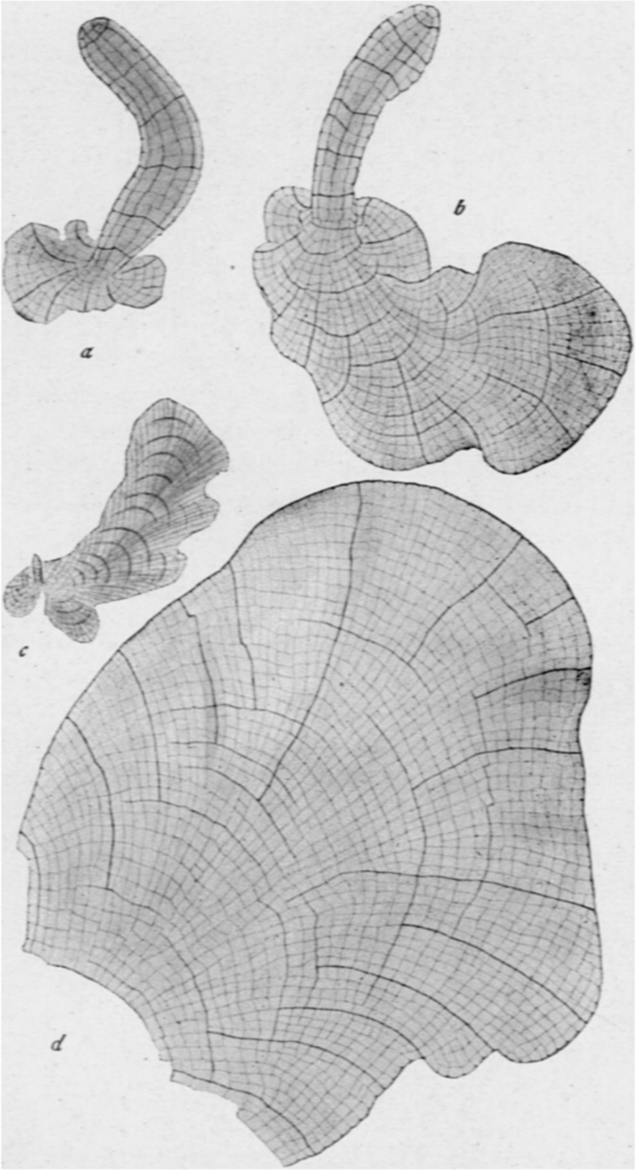


FIG. 10.—*Cutleria multifida*: normal sporelings; *a*, 20 days after fertilization; *b*, 30 days after fertilization; *d*, 55 days after fertilization (*d* represents a part of a sporeling under the same magnification as *a* and *b*, and the whole sporeling is shown in *c*); the vertical column which is the result of primary growth of the sporeling has not grown since 30 days after fertilization, and the zonal horizontal expansion at the base of the column begins as a secondary structure and develops into the characteristic *Aglaozonia* form, as shown in *b* and *c*; compare with fig. 11.

The similarity of the habit of this creeping expansion of the thallus to that of *Aglaozonia* in nature is striking. The material of *Aglaozonia* collected fresh from Posilipo and Nisida, which has grown creeping on a sea-urchin's shell or on the rock, showed a number of forms in various stage of development. If the creeping expansion of the thallus obtained in cultures is compared with *Aglaozonia* of similar size as it occurs in nature, it is hard to find any difference in their appearance. Not only the zonal growth and occasional intercalary growth of the thallus is alike in these two forms, but also the cell structure as seen in sections under the microscope is remarkably similar in the two forms. The nuclei of the thallus obtained in culture contains 48 chromosomes, and the same number is present in *Aglaozonia* as it occurs in nature.

The author's cultures of the thallus expansion, the *Aglaozonia* form of *Cutleria*, were not continued up to the stage of producing reproductive organs.

GERMINATION OF THE UNFERTILIZED FEMALE GAMETES

As was described before, the female gametes after their escape from the gametangia may swim for as long a time as 2 hours, or for as short a time as 5 minutes. When the motion becomes sluggish, the body becomes spherical, the plastids which have hitherto occupied the more posterior portion of the body become irregularly scattered throughout, and finally the cilia are either withdrawn or they coalesce with the plasma membrane as the movement ceases. If the female gametes in this condition are kept with no addition of the male gametes, they remain dormant for a considerable time. The female gamete left in this condition at length shows a change in the outermost layer of the plasma membrane that is in direct contact with sea water, the change resulting in the development of a cell wall, thin at first, and then increasing in thickness. The nucleus is in the resting condition (fig. 124). Not infrequently it is observed that the female gamete, now inclosed within a cell wall, has an elongation at a part of the body where the cell wall is thickened (figs. 125, 8, *d*).

This unfertilized female gamete commences to divide 24–28 hours after it has assumed the quiescent condition. The mitosis occurs frequently without any elongation of the sporeling. When the nucleus is in prophase there seems to be no departure from the typical mitosis characteristic of *Cutleria*, the division being perfectly normal. The number of chromosomes appearing in prophase is 24 (fig. 126). At metaphase two centrosome-like structures appear at the poles (fig. 127), and the number of chromosomes is clearly 24 (fig. 128). Sometimes the metaphase is reached while a part of the sporeling is elongating (fig. 130). The chromosomes split longitudinally and half of each chromosome passes to each pole (fig. 131). The axis of the first mitotic figure may be either longitudinal or transverse to the axis of the sporeling (figs. 132, 133). After telophase two new daughter nuclei are formed (fig. 134). In the sporeling of the unfertilized female gamete beautiful radiations are closely associated with the upper nucleus (fig. 135), but they are of short duration, and disappear completely when the nucleus passes into prophase (fig. 136). When the upper nucleus is in metaphase (fig. 137), the lower nucleus enters into prophase. Both the metaphase, viewed from pole (fig. 137), and the prophase show 24 chromosomes. One peculiarity observed is the appearance of elongated chromosomes in the upper nucleus of a 2-celled sporeling during prophase (fig. 138). The division of the upper and lower nuclei is not simultaneous; sometimes the upper nucleus is far ahead of the lower, so that the former is in late telophase

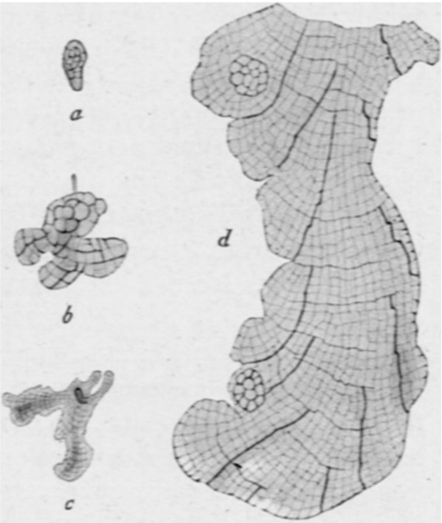


FIG. 11.—*Cutleria multifida:* apogamous sporelings: *a*, 30 days after germination; *b*, 50 days after germination; *d*, 75 days after germination (*d* represents a part of a sporeling under the same magnification as *a* and *b*, and the whole of the sporeling is shown in *c*); the apogamous sporelings are slow in development as compared with the normal ones, and assume massive forms at first and then the flat zonal expansion like the normal ones.

while the latter is in metaphase (fig. 140), or the upper is left behind in the resting state while the lower is in anaphase (fig. 139). In sporelings of the 3-celled stage the lowest nucleus may divide (figs. 141, 143) in advance of the rest, or the middle one may divide first (fig. 142).

Cell divisions continue up to a 12–22-celled stage (figs. 144–146). As seen in fig. 146, the axis of growth does not continue in one direction, as in normal fertilized sporelings, but the structure is branched. After repeated divisions there is formed a somewhat upright structure, not columnar, but rather laterally produced and in irregularly massed lumps (fig. 11, *a*). This upright structure with a lateral uneven mass consisting of about 35 cells is produced in 30 days. The irregular nature of this structure is still seen in sporelings of 50 days (fig. 11, *b*). Some of the irregular appendages of the structure which happen to be in contact with the substratum become creeping expansions and later become the main part of the sporeling.

And thus the unfertilized sporelings after 75 days develop into the thalloid creeping structure (figs. 11, *c*, *d*), and the product bears some resemblance to the product of the sporeling resulting from a fertilized gamete. Comparing these two products, the points of marked difference are as follows: (1) the first segmentation division occurs 6 hours later than in the normal sporeling; (2) the subsequent growth of these sporelings is very much slower than in normal sporelings; (3) unfertilized sporelings are characterized by developing into irregular masses at first, and even after they have assumed the expanded structure, they still show a tendency to form massed appendages; (4) the number of chromosomes is unchanged, the sporeling from the unfertilized gamete containing only 24 chromosomes.

Aglaozonia reptans

MITOSIS IN THE VEGETATIVE CELLS

Aglaozonia reptans Crouan has a creeping crustaceous habit on rock or sea-urchin's shells, attached by rhizoidal holdfasts from the superficial layer of the ventral surface. The superficial layer consisting of small cells of equal size is either single (fig. 147) or double (fig. 148) on the dorsal surface, and between the dorsal and ventral superficial layers there are two or three layers of parenchyma-like cells of huge size. Any cells in the superficial layer of the dorsal surface of young *Aglaozonia* are favorable for studying vegetative mitosis.

The vegetative mitosis was studied chiefly in the nucleus of the superficial cells of *Aglaozonia*. The main features of the division are similar to those of *Cutleria*, and consequently a detailed account and figures will be omitted at this time, but a few points should be noted.

The size of the nucleus in the superficial cells is either about the same as that of the plastids within the cell or is even smaller. When the nucleus is in the resting state, the chromatin network is remarkably similar to that of *Cutleria*, the chromatin knots being a conspicuous feature, though few in number, while the fibrils are very scanty and broken. One noteworthy feature not common in the case of *Cutleria*, but of general occurrence in *Aglaozonia*, is that deeply stained granules about the size of the chromatin knots within the nucleus are present around and close to the membrane outside the nucleus. These granules become faintly stained and evidently diminished in size during the mitotic phase within the nucleus, and they entirely disappear while the nucleus is still in prophase. It seems probable that the granules may be material allied to chromatin that might have passed into the nucleus, thus contributing to the formation of chromosomes.

The chromosomes appearing in prophase are 48 in number, and their form is in nothing different from those of *Cutleria*.

FORMATION OF ZOOSPORANGIA

Zoosporangia are produced on the upper surface of the thallus. When living *Aglaozonia* is viewed from above, the groups of zoosporangia are distinguished by patches of darker color, contrasting sharply with the light brownish olive color of the sterile portion. These patches are composed of hundreds of thousands of zoosporangia which are produced in palisade arrangement upon the thallus. The details of the origin of the zoosporangium are as follows: a superficial cell of the thallus elongates slightly and divides, giving rise to two cells, the upper one of which becomes a zoospore mother cell, and the lower remains as a stalk cell (figs. 148, 149). The process occurs in a number of superficial cells lying side by side, and as a consequence zoospore mother cells are produced in great numbers, lying close together and parallel. When the superficial layers are double, the cells in the outer layer elongate and directly become the mother cells (fig. 150). Not infrequently it is to be observed that several cell divisions take place in the superficial cells, so that a single superficial cell develops into a slender filament consisting of 3–7 cells, and the terminal one becomes the mother cell (fig. 12). The zoospore mother cell is at first like an ordinary superficial cell, with length and breadth about equal

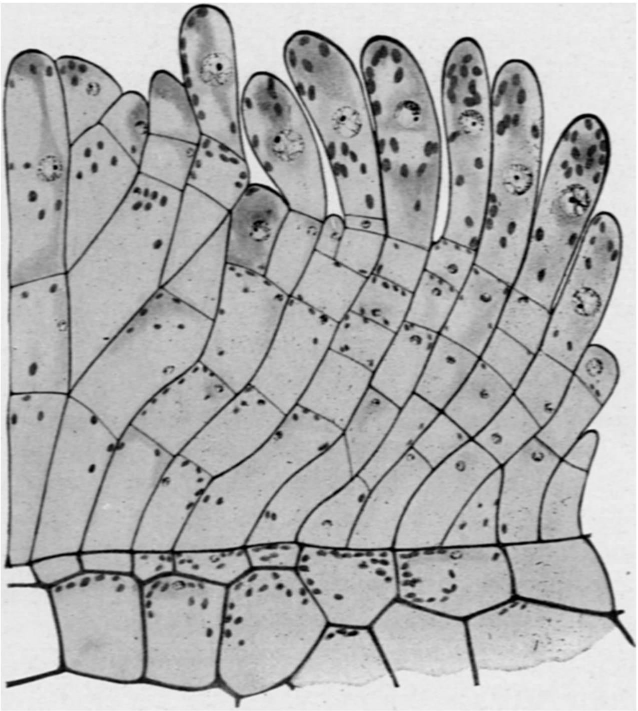


FIG. 12.—*Aglaozonia reptans*: portion of a thallus with numerous filaments; the filament usually consists of two cells, but in the present exceptional case they are made up of a varying number of cells (3–7), and their terminal cells are zoospore mother cells.

(fig. 151), but sooner or later the cell begins to elongate upward until its length becomes three times its width, the top being slightly swollen and the base narrowed. The nucleus lies either in the center or a little above the middle of the cell. The cytoplasm is full of vacuoles of various sizes, and plastids are quite numerous.

The nucleus in the resting condition contains a moderate amount of chromatin network and a nucleolus. The network is composed of very small chromatin knots and irregular fibrils (fig. 152); the proportionate amounts of the knots and fibrils seem to interchange at this stage (fig. 153). In iron-hematoxylin preparations dark kinoplasm surrounds the nucleus (figs. 151, 152). The amount of the chromatin gradually increases and the general tendency is for the broken fibrils to become thicker and continuous, transforming the knots into the fibrils (fig. 154). At this time, a single (figs. 154, 158) or double (figs. 155, 157) centrosome-like structure with radiations is recognizable at the poles.

Stronger and more continuous threads are then established by the rearrangement of the knots and fibrils. The chromatin network, which is composed of a number of broken threads of various lengths and thicknesses, now entirely devoid of former knots, is not very great in amount. For a while the broken threads are seen running for quite a distance either close to the membrane or traversing the cavity (fig. 159, 160). Then the threads gradually become more and more uniform in thickness and are transformed into somewhat regularly arranged loops, centered at a certain part of the cavity (fig. 161). This transformation represents the beginning of the synaptic stage.

This bunch of loops may be in regular arrangement (fig. 162) or adhering at the base to the membrane and diverging upward so that each loop passes along the membrane (fig. 164); or often each loop of the bunch differs in compactness of structure, and consequently some loops are short while others extend for some distance (fig. 165). In any event, a cross-section at these stages shows that the number of the cut ends of the arms of all the loops is about 48 (fig. 166). These loops now shorten considerably (fig. 167) and some of them are soon detached from the main group and form paired chromosomes (figs. 168, 169), and finally there are established 24 bivalent chromosomes, each derived from one of the loops (fig. 170). These 24 bivalent chromosomes gather near the center of the nucleus (fig. 171), and then are arranged at the equatorial plate (fig. 172), when the number is clearly counted in the polar view (fig. 173). Then each of the chromosomes splits longitudinally (fig. 174) and half of each proceeds to each pole of the spindle and there again the polar view of the anaphase clearly shows 24 chromosomes (fig. 175). The chromosomes grouped at the poles now become closely applied to one another (figs. 176, 177) and finally there are organized two daughter nuclei (fig. 178). Up to this phase a kinoplasmic mass persists, either surrounding the nucleus or near the pole of the spindle, but centrosome-like structures are recognizable only at metaphase.

The two newly formed daughter nuclei now increase in size. Their relative position within the cell is variable; sometimes they are wide apart (fig. 180), and often they lie for a while in close contact (fig. 182). The centrosome-like structure with radiations is present, associated with the two resting nuclei (fig. 180). These two nuclei may divide simultaneously or in succession (figs. 183–185). The number of chromosomes present during this division is also 24, and with this reduced number the nuclei pass into telophase (figs. 186–188). The third division follows the second after a short rest, 24 chromosomes being present at metaphase (fig. 189). As a result of the third division there are produced eight nuclei within the mother cell (fig. 190).

It is interesting to note the relative position of the axes of the mitoses that occurred within the mother cell. Only a few of the cases are represented in fig. 13. As the figures clearly show, the axis of the first mitotic figure is either in the direction of the long axis of the mother cell, or slightly oblique to the axis; in some cases the axis is still more oblique, until finally the axis of the figure is perpendicular to the long axis of the cell. In the second mitosis, considering the cases where the two mitotic figures occur at the same time, the relative position, as shown in figures, shows all possible directions of the axes. The same is true of the third nuclear division. All of these zoospore mother cells show no polarity in regard to the axis of the mitotic figures.

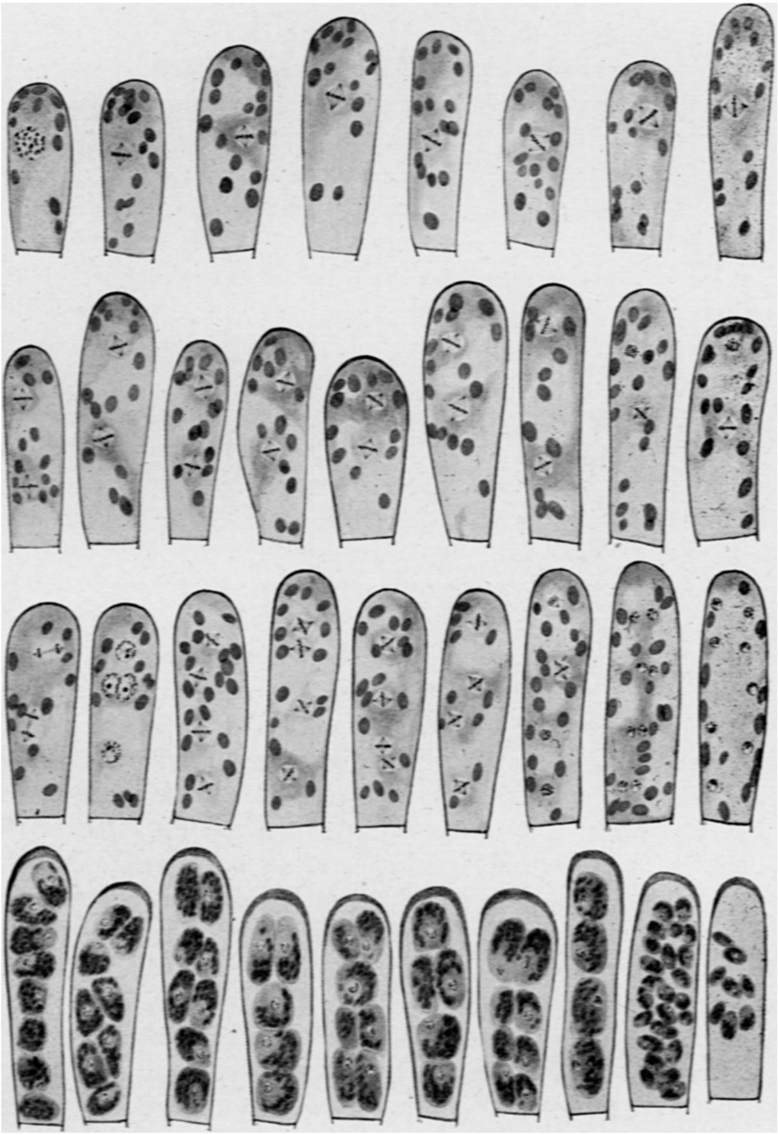


FIG. 13.—*Aglaozonia reptans:* zoospore mother cells in various stages of development; first row, the nuclear figures are in metaphase of the first division and lie in various positions in relation to the axis of the mother cell; second row, metaphase of the second division; third row, two at the right in anaphase and telophase of the second division; third row, two at the right in anaphase and telophase of the second division, two at the left in 8-nucleate stage, and the rest in metaphase of the second division; fourth row, various arrangements of zoospores formed within the mother cells, the two at the left are the two sections of a single mother cell which contains 32 zoospores, and in the rest 8 zoospores are present.

The plastids in the zoospore mother cell seem always to be multiplying in number during these nuclear divisions. When the nucleus is in prophase of the first division, the surrounding cytoplasm contains a number of plastids in various stages (fig. 191); the number is increased at the time of the 4-nucleate stage (fig. 192), and at the 8-nucleate stage the cell is filled completely with the plastids (fig. 193). It seems probable that this gradual multiplication of plastids may be one of the reasons why the zoospore mother cell constantly increases in size up to the formation of the zoospores, when the length of the mother cell becomes six times its width.

When the zoospore mother cell has reached the 8-nucleate stage, cleavage furrows generally occur in the cytoplasm, and divide the contents of the mother cell into 8 zoospore primordia (*Anlagen*). Not infrequently, however, one or two more divisions occur after the third, and as a consequence there are produced 16 or 32 nuclei, and in those cases 16 or 32 zoospores are formed (fig. 13).

The mechanism of the cleavage of the cytoplasm within the mother cell in the formation of the zoospores is noteworthy. The plastids at this time are arranged mostly near the cell membrane, and the central part of the cell is occupied by the vacuolated cytoplasm (fig. 194). The nucleus lies either near the periphery or in the midst of the cytoplasm. There is first a movement of the nuclei, and this is followed by a movement of the plastids; that is, the 8 nuclei are distributed to certain portions, not very close to the periphery, and then a number of the plastids move toward one of the nuclei as a center and surround it (fig. 195). Then there begins the separation of the *Hautschicht* from the cell wall, and at the same time cleavage furrows appear in the cytoplasm between the nuclei; the formation of the furrow is initiated by small vacuoles arranged in a cleavage line, which unite afterward and break up, leaving the furrow in their places (fig. 196). As the result of the process of the cleavage furrow, there are produced free independent zoospore primordia packed close together within the mother cell (figs. 198, 199). The relative position of the zoospores thus produced is determined by the position the nuclei had occupied before the cleavage began. Some of them are shown in fig. 13.

During the development of the zoospore mother cell, while these nuclear divisions have been taking place, the cell wall remains apparently unchanged, except at the top of the mother cell, where there has been a gradual increase in thickness, which culminates at the time of cleavage. As the zoospore primordia are completely formed, the top of the cell wall begins to swell (fig. 200), then disintegrates into lamellae, with lacunae between them (figs. 201, 202). Finally the disintegrating lamellae of the cell wall become completely disorganized, so that a pore is formed at the top, and through this the zoospores escape after maturity.

The zoospore in free swimming condition is oval (fig. 14, *a*), and usually contains more than 20 plastids. Associated with one of the plastids, which lie inside the plasma membrane, there is a red pigment spot, in close association with which two cilia of different lengths are attached, the longer one being directed toward the anterior end, about three times the length of the zoospore, and a short one extending in the opposite direction about one and one-third the length of the zoospore. The length of the zoospore is 22.5 μ when the output of the zoospores in a cell is 8 (figs. 14, *a′*, *a″*), 18.7 μ when the output is 16 (fig. 14, *b*), and 10.5 μ when the output is 32 (fig. 14, *c*).

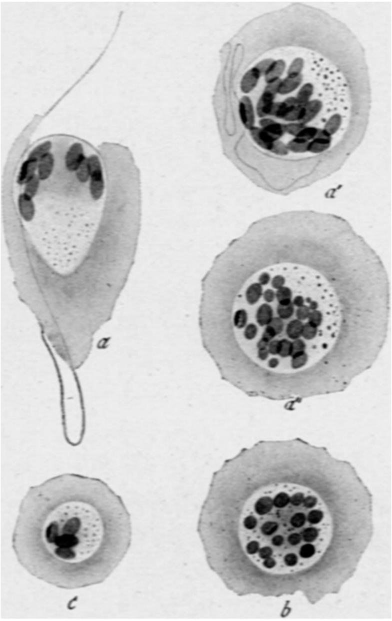


FIG. 14.—***Aglaozonia reptans:*** **zoospores sketched from living material; *a*, in free swimming condition; *a′*, beginning to be quiescent and spherical; *a″*, in quiescent condition; the size varies according to the number of nuclear divisions within the mother cell; *a*, *a′*, and *a″* are the products of three divisions (output of 8 zoospores); *b*, from four divisions (output of 16 zoospores); *c*, from five divisions (output of 32 zoospores).**

GERMINATION OF THE ZOOSPORE

The duration of the motile condition of the zoospores varies; some ceased to be motile within 10 minutes, while others were still moving after more than 90 minutes; but in any case, when the zoospore becomes sluggish, the body gradually becomes spherical and the cilia are withdrawn or become coalescent with the plasma membrane.

About two hours after the quiescence of the zoospore, the outermost layer of the plasma membrane becomes changed into a delicate cell wall. The nucleus at this time is in the resting condition, with a centrosome-like structure and radiations near the nuclear membrane (fig. 203). This condition remains unchanged during the next 20 hours (fig. 204). The presence of centrosome-like structures and radiations is not a definite feature; at about 24 hours after quiescence they may (figs. 206, 207) or may not be visible (figs. 205, 208).

The first segmentation division of the germinating zoospore takes place 24 hours after its quiescence. The nucleus enters prophase either at the center of the cell (fig. 209) or in its elongated part (fig. 210), and when the chromosomes are arranged at the equatorial plate (figs. 211, 213), two centrosome-like structures are present at the poles. The polar view of the metaphase shows distinctly 24 chromosomes (figs. 212, 214). Each of the chromosomes divides longitudinally and half of each passes to each pole (fig. 215). The second mitosis in these two nuclei may occur in succession (figs. 217–221) or simultaneously (fig. 222), but in either case 24 chromosomes can be counted at prophase and in polar view of metaphase.

In the zoosporelings consisting of three cells the three nuclei divide in various order (figs. 224, 225), and in prophase of any nucleus 24 chromosomes are present. In the further nuclear divisions in the development of the zoosporelings the same number of chromosomes is counted. This young sporeling after a multitude of cell divisions (figs. 226–228) develops into a long filamentous structure instead of a stout column. The product at about the end of 20 days after germination is shown in fig. 15, *a*. In habit the sporeling is very like the young filamentous stage of *Cutleria*.

Later this filament does not continue in the upward direction, but there appears a new lateral structure near the basal region of the primary filament, and the whole structure clings to the substratum with rhizoids which grow from the base; thus, contrary to the behavior of the fertilized gamete of *Cutleria*, basal expansions that creep flat upon the substratum are never found. The plant at 30 days is illustrated in fig. 15, *b*, which resembles the young *Cutleria* as found in nature.

The new structure thus initiated laterally from the basal region of the primary filament grows in such a way that it finally meets the other side of the primary filament so as to form a funnel which is expanded upward and narrowly constricted downward. Upon the expanded upper margin of this funnel-shaped structure delicate hairs begin to grow. This stage is reached in about 40 days (fig. 15, *c*). These funnel-shaped structures obtained in the artificial culture present a striking resemblance to the young plants of *Cutleria* in nature as they occur in tufts on the rock or on shells. One difference is that the young plant of *Cutleria* in nature, at the same stage as the cultures in the

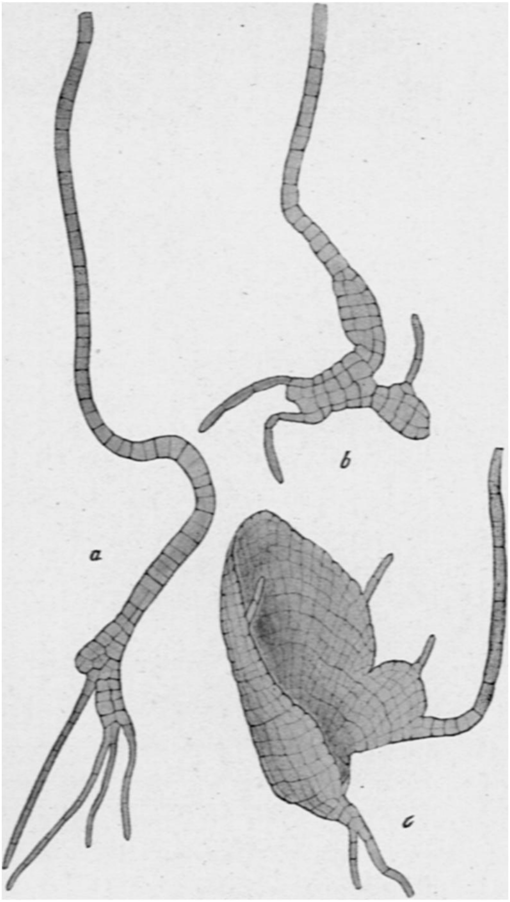


Fig. 15.—*Aglaozonia reptans:* zoosporelings; *a*, 20 days after germination; *b*, 30 days after germination; *c*, 40 days after germination; the sporelings are filamentous in primary growth as shown in *a*, then gradually assume the funnel-shape shown in *b* and *c*, this shape being characteristic of the young stage of *Cutleria* in nature.

laboratory, is furnished with a luxurious display of long beautiful hairs on the margin of its funnel-shaped thallus, while such a hairy growth has not yet been seen in cultures. It must be admitted, however, that there is a difference between conditions in nature and in cultures which may account for the difference in this character; the young plant of *Cutleria* is found on the rock at a depth of 1–5 meters, while the culture was kept in a tank where the water was never deeper than 6–8 inches. Intensity of light, temperature of water, its pressure, its movement, and other factors may be quite different, and yet in spite of such a difference of external environment, the zoosporelings of *Aglaozonia* developed into erect structures of funnel-shape as in the young *Cutleria* plants, and fundamentally different from the flat creeping expansion habit of both parent forms and of the plants resulting from fertilized female gametes of *Cutleria*.

The young plants of *Cutleria* and the product of the zoosporelings of *Aglaozonia* in culture show not only the common habit in their development and similarity in their outer morphological characters, but also a similar cell structure, similar nuclear conditions, and the same number of chromosomes.

Discussion of cytological phenomena

THE ORIGIN OF UNIVALENT CHROMOSOMES

The chief constituents of the resting nucleus of *Cutleria* and *Aglaozonia* are the network and nucleolus. The network is composed of two parts, granules and irregular fibrils in varying proportions. The granules are of different sizes and some of them are connected by very slender, irregular fibrils, or lie isolated usually near the nuclear membrane. The number of granules in the nucleus quite often, though not always, corresponds more or less to the number of chromosomes. The granules may be extremely small, but the preparations never fail to show their presence. Both granules and fibrils consist of chromatin.

As a rule, there is only one nucleolus, and it lies quite free from the nuclear network, with which it shows no visible physical connection. Concerning the morphological function of the nucleolus in algae, STRASBURGER (**55, 57**) published the view that the substance of the nucleolus is utilized for spindle formation, the conclusion being drawn from the fact that the nucleolus disappears partly or completely immediately preceding the formation of the spindle. WILLIAMS (**66**) in his study of *Dictyota* accepted this view. STRASBURGER (**53**) had held the opinion that the nucleolus was reserve material serving to build up the chromosomes, and this view was followed by SWINGLE (**61**) on *Stypocaulon* and by MOTTIER (**32**) on *Dictyota*. The view that the chromosomes are formed directly from the nucleolus was supported by TANGL (**62**), MEUNIER (**29**), MOLL (**30**), DECAGNY (**10**), HENNEGUY (**19**), VAN WISSELINGH (**67–69**), BERGHS (**3**), KARSTEN (**22**), and TRÖNDLE (**64**) in their studies of *Spirogyra*, by WOLFE (**70**) on *Nemalion*, by LEWIS (**27**) on *Griffithsia*, and by SVEDELIUS (**60**) on *Delesseria*. These investigations differ greatly in regard to exactness and details. In some of these papers views are expressed with adequate illustrations, but often the illustrations and text are quite contradictory, and frequently the figures would afford better support to some other interpretation. In their statements, however, these authors seem to agree that some or all of the chromosomes are organized directly from the material of the nucleolus, which in this way either may be used up partly or even entirely.

A fourth opinion is that the chromosomes arise exclusively from the chromatin network, the nucleolus taking no direct part in their development. *Fucus* illustrates the situation. The plates accompanying the studies on *Fucus* by STRASBURGER (**56**), and by FARMER and WILLIAMS (**15**) show this situation, and more details have been furnished by the present author (**73**). *Polysiphonia* (YAMANOUCHI **71**), *Corallina* (DAVIS **8**, and a forthcoming paper by the author), *Zanardinia* (YAMANOUCHI **76**), as well as *Cutleria* and *Aglaozonia*, present a similar situation.

THE ORIGIN OF BIVALENT CHROMOSOMES

Contrasted with the formation of univalent chromosomes in typical mitosis, the origin of bivalent chromosomes is rather complex. As already described, in the resting nucleus of the zoospore mother cell of *Aglaozonia* the chromatin network is composed of granules and fibrils. While in typical mitosis the network finally becomes transformed into isolated individual chromosomes, the granules and fibrils being entirely used up in the formation, in the reduction mitosis the granular parts become transformed into fibrils, and consequently during the presynaptic stage there are only thread structures within the nucleus. These chromatin threads, at their beginning irregularly thickened and branched, become much evener, and the transformation continues until long continuous threads are formed, which run freely throughout the cavity. The threads thus formed, from the beginning of their transformation to their completion as continuous structures, have a single nature. Entering the synaptic condition, the single threads then shorten and thicken, and become either eccentrically grouped as a loose tangled mass at one side of the nuclear cavity, or are variously scattered all over the membrane. The threads eventually form loops by repeated folding. The number of loops is 24. Each loop folds together at its bent end so that the bent arms come into contact with each other in the culmination of synapsis. As they emerge from synapsis, there are present 24 bivalent chromosomes, which become detached from the nuclear membrane, moving toward various parts of the nuclear cavity.

The relationship of the chromatin threads in prophase, the loops in synapsis, and the bivalent chromosomes of postsynapsis have been clearly traced. A pair of bivalent chromosomes corresponds to one of the loops in synapsis, the loop being formed by a folding back of the chromatin thread, so that a loop in synapsis should be considered as composed of two sporophytic chromosomes associated end to end. The two elements of the bivalent chromosomes separate from each other at the metaphase of the first reduction division, thus effecting what may be regarded as a qualitative reduction.

The origin and behavior of bivalent chromosomes as described for *Fucus* (**73**) and *Zanardinia* (**76**) agree perfectly in essentials with the present account of *Aglaozonia*. For example, figs. 161, 162, and 172, illustrating looping threads in synapsis and chromosomes in late prophase in the zoospore mother cell of *Aglaozonia*, when compared with figs. 43, 44, and 49 illustrating similar stages of *Fucus* (**73**), demonstrate at once their similarities.

Disregarding many points which may differ in particulars, the case of *Aglaozonia*, in which bivalent chromosomes in seemingly reduced number have originated by the folding back of sporophytic chromosomes united end to end, will agree in essentials with observations made by different authors on various forms, for example, by FARMER and MOORE (13, 14), SCHAFFNER (52), and MOTTIER (34) on *Lilium*, by STRASBURGER (58) on *Galtonia*, and by one group of zoologists, such as VON RATH (37) on *Gryllotalpa*, RÜCKERT (41) on copepods, and MONTGOMERY (31) on *Peripatus*.

In general, while the method of forming bivalent chromosomes by the end to end fusion of sporophytic chromosomes or *métasyndèse*, as GRÉGOIRE (17) calls it, occurs in *Fucus*, *Zanardinia*, *Aglaozonia*, and other forms, there is also another method by side to side fusion, or *parasyndèse*, in *Polysiphonia* (71), *Nephrodium* (72), *Osmunda* (75), and various other forms, such as *Lilium* (GRÉGOIRE 16, BERGHS 2, ALLEN 1), *Allium* (BERGHS 2), *Trillium* (GRÉGOIRE 16), and *Drosera* (ROSENBERG 40). From these observations it is evident that in the former class there is no duality in the formation of thread structures (*leptotènes*) from the resting nucleus, and therefore in synapsis the two bent arms of a single loop come into close association; and in the latter class, a duality is present at the time of the beginning of the thread structures (*leptotènes*), and the two independent members of the duality come into contact during synapsis. In both classes, each of the two elements of the bivalent chromosome is derived from a single sporophytic chromosome, and the two elements separate in the first division. It seems highly probable that there are two distinct types of arrangement of sporophytic chromosomes at synapsis in plant cells.

POLAR ORGANIZATION

Views on the polar organization of plant cells are conflicting. Some of the algae present clear evidence of such polarity, the best known examples being *Stypocaulon* and *Sphacelaria* (61), which have a center in the form of an aster with a centrosome at the side of the resting nucleus of the apical cell. The centrosome is said to divide previous to each mitosis, to establish the poles of the spindle. A similar centrosome with radiations is present in the tetraspore mother cell of *Dictyota* (**32, 66**). Other algae, such as *Corallina* (**8**), *Fucus* (**15, 56, 73**), *Polysiphonia* (**71**), show highly advanced centrospheres at the poles of the spindle; but investigators claim that in *Corallina* and *Polysiphonia* they appear simultaneously at the two poles during prophase, culminate at metaphase, and disappear at anaphase or telophase; and in *Fucus* one of them appears before the other and they disappear at telophase; only as rare cases they remain recognizable at the side of the resting nucleus. Thus, so far as present knowledge goes, the persistent continuity of polar organization in algae cells has not been demonstrated through successive cell generations.

The subject of polar organization has received especial attention in HARPER'S studies on mildews, especially *Phyllactinia* (**18**). In this form, the "central body" lies within the membrane of the resting nucleus, and it is connected with chromatin strands so as to give polarity to the nucleus. The poles of the spindle are formed by division of the central body. HARPER believes in the permanence of this structure from mitosis to mitosis, and in the persistence of its connection with chromatin.

The studies of recent years on the cells of pteridophytes and spermatophytes have failed to support certain claims for the presence of centrosomes and have indicated that their cells are without visible polar organization. MARQUETTE (**28**) published the view that the presence of polarity in the leaf cell of *Isoetes* is manifested by a large starch-containing body that lies closely pressed against the side of the nucleus. Previous to nuclear division, according to MARQUETTE, the polar structure divides, and during mitosis the structure persists up to telophase, and finally one of them remains at the side of the newly formed daughter nucleus, and a similar constriction division occurs in the structure previous to the next nuclear division. He also found a polar organization of the spore mother cell of *Marsilia*, in the form of aggregated starch grains, conspicuous at the time of synapsis, but disappearing just before the formation of the first spindle; the organization is of very short duration, not continuing throughout the mitosis.

In *Cutleria* and *Aglaozonia*, polar organization is manifested by the appearance of centrosome-like structures with or without radiations, always only at metaphase, and as a rule completely disappearing at telophase. It may be found that polarity does not depend in all cases upon the presence of permanent protoplasmic structures of recognizable size or upon such morphological differentiation as to rank as organs of the cell. There may possibly be polarity at times without visible protoplasmic organization, but so far as the visible organization of the cell is concerned, there seems to be no permanent polar organization in the cells of *Cutleria* and *Aglaozonia*. The transient polar organization is formed *de novo* at each mitosis in every possible position about the nucleus. The variability of the polar axis of the mitotic figure demonstrates the feature (fig. 13).

Zoospores and gametes of algae invariably present a conspicuous polarity in that their cilia are situated at one end or at a definite point on the side, the point of insertion being devoid of plastids. The most vital problem of zoospore and gamete formation is whether the polar organization of these cells arises *de novo* at the time of development or is handed down from the succession of the cells that are their progenitors. STRASBURGER (**57**), from his studies of the zoospores of *Oedogonium*, *Cladophora*, and *Vaucheria*, decided that the cilia bearing organ, the blepharoplast, arises in the plasma membrane (*Hautschicht*), the nucleus lying in close association at the time of its formation. MOTTIER (**33**) later described a similar origin of the blepharoplast in *Chara*. DAVIS' work on *Derbesia* (**9**) showed that the blepharoplast is developed at the plasma membrane in the form of a ring that has been organized by the side by side union of numerous granules arranged in a circle. The marked polarity is manifested after the appearance of the granules.

The details of the organization of zoospores and gametes of *Cutleria* and *Aglaozonia* have not been given in this account because the subject will be dealt with in a special paper treating of the origin of motile sperms or spores of algae, such as *Fucus*, *Zanardinia*, *Ectocarpus*, and of some green algae, so that a brief account will be sufficient for the present consideration of polarity.

In the zoospore mother cell stage, which is usually reached at the 8-nucleate stage of the zoosporangium, cleavage furrows start from the periphery and cut into the protoplasm in the form of curved and branching furrows. Finally, the protoplast within the sporangium becomes divided into approximately equal masses, each of which contains a single nucleus. These masses are zoospore primordia, and each develops into a uninucleate zoospore. The nucleus lies first at the center of the zoospore primordium, with plastids lying near the periphery of the cell. The polarity of the cell is clearly established when the nucleus moves toward the peripheral region of the cell, displacing all the plastids in the vicinity, except one which remains near the nucleus. At this time a small granule can be seen lying in the plasma membrane near the nucleus. This granule becomes the blepharoplast, and in a part of the body of the plastid remaining near the nucleus there is later developed a red pigment which makes it look as if the cilia had arisen from the red pigment.

Thus the polarity of the zoospore is manifested only after the organization of the blepharoplast and red pigment spot in the plastid. Again, after its quiescence, the exhibition of polarity is lost until either the segmentation of the nucleus or the elongation of the cell wall begins.

ALTERNATION OF GENERATIONS

Since the classical investigations of HOFMEISTER (**20**), it has been universally recognized that the Archegoniatae are characterized by a definite alternation of generations. In this group there are two regularly alternating generations, one bearing sexual reproductive organs and the other producing spores. In sharp contrast to the unanimous agreement in regard to the existence of an alternation of generations in the Archegoniatae, there is a lively contest of contradictory views as to the alternation of generations in the thallophytes. The thallophytes being a loose assemblage of widely diversified types, and investigations on their life histories being far from numerous enough to permit of any generalization, some still question whether a regular alternation of definitely established generations exists in the group, and if it exists, whether it is to be regarded as homologous or antithetic.

Perhaps the first clear statement of a regular alternation of generations in thallophytes is by SACHS (42), who endeavored to bring together the facts known in algae and fungi, and to compare them with alternation in the Archegoniatae. SACHS states that the life cycle of algae and fungi is similar to that of the Archegoniatae, one generation producing sexual organs, and the other forming spores. PRINGSHEIM (36) held quite a different view, namely that the alternation of generations in the thallophytes consists in the regular succession of a non-sexual or "neutral" generation with a sexual generation, both generations being of similar structure.

Both these views, one by SACHS, who recognized two distinct generations in thallophytes, and the other by PRINGSHEIM, who regarded the two alternating generations as similar structures, have continued to find followers. VINES (65) held the view that most of the thallophytes have no alternation of generations, since both sexual and asexual modes of propagation are directly dependent upon the external conditions, and that an alternation of generations in algae comparable to that in the bryophytes is only found in *Coleochaete* and *Chara*. CELAKOVSKY (5), although opposing PRINGSHEIM in his conception of alternation of generations in the Archegoniatae, which he designated as the "antithetic," agreed with him in his conception of alternation in thallophytes, in which the successive generations are alike and which conception he designated as the "homologous." PRINGSHEIM'S conception of the homologous alternation of generations in the Archegoniatae has received its principal support in LANG'S experimental cultures (26) of the apogamous development of the sporophytes on prothallia of several pteridophytes. CELAKOVSKY'S conception of alternation of generations in the Archegoniatae was taken up fifteen years later by BOWER (4), who has supported this conception by his theory of an interpolated sporophytic generation. BOWER holds that the antithetic alternation originated by the intercalation of a non-sexual generation as a new development between two gametophytic generations. This interpolation of a special sporophyte probably took place, according to BOWER, in the algae-like ancestors of the Archegoniatae as they emerged from an aquatic life to a land life. Moreover, some of the thallophytes, such as *Coleochaete*, Ascomycetes, and Florideae, he holds show the beginning of an antithetic alternation. STRASBURGER (**54**), summarizing cytological results, advanced the theoretical conception of the periodic reduction of chromosomes in the life cycle in plants, thus establishing the view that the x and $2x$ generations which complete the life cycle of the plant are quite distinct from each other. This view has proved the antithetic character of the two generations from a cytological standpoint.

Since that time, cytological investigations of algae which directly touch the problem of the alternation of generations are those of KLEBS (**23**), WILLIAMS (**66**), WOLFE (**70**), LEWIS (**27**), SVEDELIUS (**60**), and of the author (**71, 73, 74, 76**).

KLEBS conducted experimental cultures of several algae, such as *Vaucheria*, *Oedogonium*, and others. By controlling factors such as light, temperature, moisture, oxygen, and chemical composition of nutritive media, he succeeded in producing any kind of reproduction, either sexual or asexual; in other words, he observed no regular alternation of neutral and sexual generations.

WILLIAMS discovered a reduction of chromosomes during the tetraspore formation in *Dictyota*, and this led him to conclude that the tetrasporic plant of *Dictyota* is a sporophytic generation derived from the fertilized egg.

WOLFE showed for *Nemalion* that the cells of the cystocarp have double the number of chromosomes found in the sexual plant, thus presenting the first cytological evidence that the cystocarp of the red algae is sporophytic in character. He placed the period of chromosome reduction at the time of carpospore formation, basing his conclusion on a count of chromosomes in the mitosis just previous to the formation of the carpospores. However, he did not describe the phenomena characteristic of chromosome reduction, namely, the period of synapsis followed by the two divisions which distribute the chromosomes so as to give a numerical reduction.

After the publication of WILLIAMS' and WOLFE'S work, STRASBURGER expressed his agreement with WILLIAMS' conclusion concerning the alternation of generations in the brown algae, but remarked that the tetraspores of the red algae seemed to be different from those of the Dictyotaceae, and that the place of the chromosome reduction in the red algae should be sought elsewhere than at the formation of tetraspores because some of the red algae develop no tetraspores, but monospores instead. But immediately after the appearance of this view of STRASBURGER, the author's work on *Polysiphonia* was published. He found that the reduction of chromosomes, instead of taking place at the formation of carpospores as reported by WOLFE, occurred at the development of the tetraspores. Thus in the life cycle of *Polysiphonia*, the sporophytic generation is not only represented by cystocarpic branches, but also extends to the tetrasporic plant that alternates antithetically with the sexual plant.

LEWIS then worked out the cytology of *Griffithsia*, special attention being paid to the problem of the alternation of generations. The place of the reduction of chromosomes is in tetraspore formation as in *Polysiphonia*. He concludes that there is in *Griffithsia* as in *Polysiphonia* an antithetic alternation of generations, the gametophytes being represented by the sexual plants and the sporophytes by the sporogenous cells of the cystocarps. In addition to this, there is a regular succession of tetrasporic individuals and sexual individuals; tetrasporic individuals resemble the gametophytes in morphological differentiation and the sporophytes in number of chromosomes. He regards the tetrasporic and sexual plants as presenting homologous alternation.

SVEDELIUS studied the alternation of generations in *Delesseria*. Although his work does not cover the whole life cycle of this type, the cytological studies being made only in the development of tetraspores and vegetative mitosis in both tetrasporic and female plants, yet by comparing his work with that on *Nemalion*, *Polysiphonia*, and *Griffithsia*, he has come to the conclusion that there is an alternation of generations in *Delesseria* as in *Polysiphonia*.

The existence of an alternation of generations in the thallophytes, though somewhat obscure in the green algae on account of insufficient investigations, has been clearly proved by the cytological studies on *Fucus* and *Dictyota* among the brown algae, and on *Nemalion*, *Polysiphonia*, *Griffithsia*, and *Delesseria* among the red algae. Let us now turn to the situation in *Cutleria*.

Since FALKENBERG (12) first suggested that *Cutleria* and *Aglaozonia* may represent the two phases of the life cycle of a single species (*Cutleria*), many workers have tried to secure *Aglaozonia* plants from the cultures of fertilized gametes of *Cutleria*. FALKENBERG and REINKE were the only two who succeeded in producing the form *Falkenbergiana*, an *Aglaozonia* form. The rest of the investigators succeeded in producing either the form *Falkenbergiana* or the form *Churchiana* from unfertilized gametes. Among later workers, SAUVAGEAU, by carrying on his cultures for years, succeeded in getting *Aglaozonia* or *Cutleria* from unfertilized gametes of *Cutleria adspersa*. The conclusion drawn from this result naturally was that the alternation of generations is not necessary, but rather, as it might be called, facultative. OLTMANNS and STRASBURGER have agreed with this view of SAUVAGEAU. The so-called polymorphic character of *Cutleria*, however, needs analytical consideration. The results of the author's investigation are as follows:

The investigation of *Cutleria* plants bearing gametangia showed that the nucleus has 24 chromosomes in both the vegetative and germ cells. The number is doubled at fertilization by the union of the sexual nuclei. The *Cutleria* plant is therefore a gametophyte whose chromosome number is 24. As all now agree, the fertilized gamete or egg with its fusion nucleus should represent the $2x$-generation, and so the fertilized gamete of *Cutleria* is the beginning of the $2x$-generation. The studies of the germination of the fertilized gamete have shown that the $2x$ condition continues in further development. The sporeling developed into a structure unlike the young filamentous form of the parent *Cutleria* as found in nature, but composed of a small upright column and a proportionally large basal disk expansion which became later the dominating region of the further development. Later this expansion develops the zonal structure of *Aglaozonia* as found in nature, and not only resembles *Aglaozonia* in outer morphology, but also in the nuclear conditions, the expansion in cultures and *Aglaozonia* in nature both having 48 chromosomes. *Aglaozonia* in nature produces zoospores, during whose formation 48 chromosomes are reduced to 24, and the zoospores contain 24 chromosomes. The zoospore with the haploid number germinates with no fusion. The product of the zoosporeling is a filamentous individual like young *Cutleria*, and never produces the zonal flattened form of the parent *Aglaozonia*.

The condition in cultures is very different from the environment in nature. In nature the gametes and zoospores are set free at a depth of 1–5 meters and in cultures they are discharged and kept in sea-water at a depth less than 15–20 cm. Besides, the intensity of light, water-pressure, temperature, and motion of water are also different in these two different environments, and yet the sporelings of the fertilized gametes developed into the flattened disk like young *Aglaozonia* and those of the zoospores grow into filamentous plants like young *Cutleria*. That the potentialities of those two kinds of sporelings in forming invariably the particular individuals different from the parent forms as in nature, even under environmental conditions so different from what is found in nature, and that the potentialities of the two different sporelings have given rise to the particular different individuals, even when they were kept in similar cultures, show that the potential characters of their germ plasm has dominated over the influence of environmental conditions; this dominancy of the innate character over the environmental influence is fundamentally different from the experiments of KLEBS on certain green algae (**23**). Although the two kinds of sporelings from fertilized gametes and zoospores in the cultures have not been kept growing to the stage of reproduction, yet it seems safe enough to infer that the disklike expansion developed from the sporelings of the fertilized gamete will be identical with *Aglaozonia* as found in nature, and the filamentous structures of the zoosporelings will be *Cutleria*. Thus *Cutleria* plants with zoospores represent a gametophytic generation, and *Aglaozonia* plants with fertilized gametes the sporophytic generation. These two generations alternate in the life history of *Cutleria*. The two generations, from the above observations of the cell organization, are fundamentally different and could not be regarded as an example of polymorphism in the sense of OLTMANNS (**35**).

The observations on the sporelings of the unfertilized gametes and studies of their cell organization undoubtedly show that the female gametes of *Cutleria* are capable of developing into the flattened disk like *Aglaozonia* in nature. Though at first their external morphology presents irregular deviations from forms in nature, the nuclear divisions are perfectly normal. The culture of the sporelings was not continued up to production of reproductive organs, and consequently the discussion cannot be carried on any further. This apogamy may indeed be a reversion to the ancestral type of asexual spores which certainly existed before the appearance of sexuality in the gametophytic generation, but when sexuality is once established, the fusion of gametes and thereby the sporophytic generation is interpolated as a secondary structure in which the reduction of chromosomes occurs.

Summary

The nuclear conditions in the life history of *Cutleria multifida* may be summarized as follows:

1. The nucleus of both male and female plants contains 24 chromosomes, and the male and female gametes contain the same number.

2. In the union of gametes the number is doubled, and 48 chromosomes appear in the sporelings, which develop into the *Aglaozonia* form of *Cutleria*. Therefore, the individual bearing the name of *Cutleria multifida* represents the gametophytic phase of the species, 24 being the gametophytic number of chromosomes; and the *Aglaozonia* form of *Cutleria* represents the sporophytic phase, 48 being the sporophytic number.

3. *Aglaozonia reptans* contains 48 chromosomes, and the number is reduced in zoospore formation, the zoospore containing 24 chromosomes. The zoospore with the reduced number of chromosomes germinates without conjugation. The individual grown from the germinating zoospore presents a striking similarity to the young form of *Cutleria* in nature and contains 24 chromosomes, the same number as the latter. Therefore it is evident that the individual bearing the name of *Aglaozonia reptans* represents the sporophytic phase of the species, 48 being the sporophytic number of chromosomes, and that the gametophytic phase is represented

by the individual grown from the zoospore and resembling the young form of *Cutleria* in nature. It is certain that *Aglaozonia reptans* as it occurs in nature is identical with the *Aglaozonia* form of *Cutleria multifida* as developed in cultures and now determined to be the sporophytic phase of the species.

4. Therefore, *Cutleria multifida* and *Aglaozonia reptans* represent respectively gametophytic and sporophytic generations of a single species, the two generations alternating in the life history of *Cutleria*.

5. The female gamete of *Cutleria* may germinate apogamously. There is no irregularity in the mitotic process, 24 chromosomes being invariably present. The individual produced, in its early development differs somewhat from the product of the fertilized gamete, but the fate of the apogamous individual was not determined.

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EXPLANATION OF PLATES XXVI–XXXV

All figures were drawn with the aid of a camera lucida under the Zeiss apochromatic objective 1.5 mm. N.A. 130, in combination with compensating ocular 12; except figs. 200–202, which were drawn with compensating ocular 18; figs. 8, 9, 14, drawn with compensating ocular 8; and figs. 1–7, 12, 13, 16, 17, 53, 144–150, drawn with ocular 4; and figs. 10, 11, 15, which were drawn under Zeiss apochromatic objective 16 mm. combined with compensating ocular 12. The figures are reduced to one-half the original size. Figs. 1–15 are in the text.

PLATE XXVI

The formation of the male gametangium of Cutleria multifida

FIG. 16.—Portion of the thallus of a male plant, showing a single layer of superficial cells and the larger cells below.

FIG. 17.—Portion of the thallus of a male plant, where 4 superficial cells have grown into papillae, the male gametangium initials.

FIG. 18.—Male gametangium initial: nucleus in resting condition.

FIG. 19.—Prophase of nucleus: 24 chromosomes and a nucleolus present.

FIG. 20.—Metaphase: the nuclear cavity in this stage strikingly diminished.

FIG. 21.—A filament with two male gametangia: nucleus in the young gametangium initial at metaphase.

FIG. 22.—Anaphase that follows the metaphase of fig. 20.

FIG. 23.—Nucleus already divided and each daughter nucleus now in prophase; 24 chromosomes present in each.

FIG. 24.—Nucleus in the lower cell in advanced prophase, while the nucleus in the upper cell is still in early prophase.

FIG. 25.—Similar stage to fig. 24, excepting the presence of centrosome-like structures at the poles.

FIG. 26.—Nucleus in the lower cell in anaphase; nucleus in the upper cell in resting condition.

FIG. 27.—Metaphase of the nucleus in the upper cell of 2-celled stage; the lower cell is not drawn.

FIG. 28.—Late metaphase of the nucleus in the upper cell; nucleus in the lower cell in resting condition.

FIG. 29.—Telophase of nucleus of the upper cell; a new cell wall is being laid down between two daughter nuclei.

FIG. 30.—Male gametangium of 3 cells: the nuclei in the upper two cells are in resting condition; the nucleus in the basal cell in prophase, showing 24 chromosomes and a nucleolus.

FIG. 31.—Male gametangium of 3 cells: the nucleus in the terminal cell is in metaphase; centrosome-like structures at the poles of the spindle.

FIG. 32.—Male gametangium of 3 cells: the nucleus in the terminal cell in metaphase, with centrosome-like structures at the poles; the nucleus in the middle cell in anaphase, and that of the basal cell in resting stage.

FIG. 33.—Male gametangium of 4 cells: the nucleus in the terminal cell is in metaphase; that of the middle cell in prophase, showing 24 chromosomes.

FIG. 34.—Male gametangium of 4 cells: two nuclei are in prophase; 24 chromosomes are clearly present; one in anaphase and the other in resting condition.

FIGS. 35, 36.—Mitotic figures of the nucleus in a vegetative filament: fig. 35, anaphase; fig. 36, prophase, showing 24 chromosomes.

FIGS. 37, 38.—Cross-section of a male gametangium of 5 cells: fig. 37, metaphase of the mitosis which divides the filament into two rows of cells; fig. 38, prophase of the same, showing 24 chromosomes.

FIG. 39.—Tip of a vegetative filament, showing stages in the formation of male gametangia; the male gametangia have arisen from the third, fourth, sixth, and seventh cells; four nuclei in the three young gametangia are in prophase; 24 chromosomes can be counted in each.

FIG. 40.—Portion of a male gametangium of 2 rows of cells: one nucleus in metaphase, one in prophase showing 24 chromosomes, and three others in the resting condition.

FIG. 41.—Portion of a more advanced male gametangium: four nuclei in prophase, clearly showing the 24 chromosomes.

FIG. 42.—Cross-section of a male gametangium at about the same stage as in fig. 41; one nucleus is in prophase; 24 chromosomes present.

FIG. 43.—Portion of a male gametangium at about the same stage as in fig. 41; two nuclei are in anaphase.

FIG. 44.—Cross-section of a male gametangium at about the same stage as in fig. 43: two nuclei in prophase of the mitosis which divides the filament into a structure with four rows of cells.

FIG. 45.—Portion of a male gametangium consisting of 4 rows of cells: one nucleus is in anaphase.

FIG. 46.—Portion of a male gametangium at about the same stage as in fig. 45: one nucleus is in prophase, showing clearly 24 chromosomes.

FIGS. 47, 48.—Portion of a male gametangium at a more advanced stage: one nucleus in fig. 47 in prophase, showing 24 chromosomes; one nucleus both in figs. 47 and 48 in metaphase; and two nuclei in fig. 48 in anaphase.

FIG. 49.—Portion of a male gametangium, showing the last division of nuclei; one nucleus is in prophase and the other in metaphase.

FIG. 50.—Cross-section of a male gametangium at about the same stage as in fig. 49; one nucleus is in prophase, two others in metaphase of the mitosis which divides the filament into a structure of 8 rows of cells.

FIG. 51.—Cross-section of a mature male gametangium: nuclei are in resting condition.

FIG. 52.—Cross-section of a mature male gametangium, showing the plastids that are associated with the red pigment.

PLATE XXVII

The formation of the female gametangium of Cutleria multifida

FIG. 53.—Portion of thallus of a female plant, showing a single layer of superficial cells and the larger cells below; one of the superficial cells has increased in size, and will be the female gametangium later.

FIG. 54.—One of the superficial cells becomes still larger in size: nucleus is in prophase; 24 chromosomes and a nucleolus are present.

FIG. 55.—Nucleus in metaphase; centrosome-like structures at the poles of spindle.

FIG. 56.—Anaphase.

FIG. 57.—Later telophase: cell wall is being laid down between the newly formed daughter nuclei.

FIG. 58.—Female gametangium of 2 cells.

FIG. 59.—Nucleus of the upper cell in prophase; nucleus of the basal cell in resting condition.

FIG. 60.—Nucleus of upper cell in metaphase.

FIG. 61.—Nucleus of upper cell in anaphase.

FIG. 62.—Late telophase of nucleus in upper cell.

FIG. 63.—Female gametangium consisting of 2 cells: nucleus of basal cell in metaphase.

FIG. 64.—Nucleus in basal cell in anaphase.

FIG. 65.—Female gametangium of 3 cells: nucleus of basal cell in prophase; 24 chromosomes and a nucleolus are present.

FIG. 66.—Nucleus of terminal cell in prophase, showing 24 chromosomes; nucleus of basal cell in metaphase.

FIG. 67.—Nucleus of terminal cell in metaphase.

FIG. 68.—Nucleus of terminal cell in anaphase.

FIG. 69.—Nucleus of middle cell in prophase.

FIG. 70.—Nucleus of middle cell in early anaphase.

FIG. 71.—Cross-section of a female gametangium of 2 cells: nucleus in anaphase viewed from the pole, clearly showing 24 chromosomes.

FIG. 72.—Cross-section of a female gametangium of 5 cells: nucleus in metaphase of the mitosis which divides the gametangium into a structure composed of 2 rows of cells.

PLATE XXVIII

The formation of the female gametangium of Cutleria multifida (continued)

FIG. 73.—Cross-section of a female gametangium of 8 cells in two rows: one nucleus in late prophase, showing 24 chromosomes.

FIG. 74.—Portion of a female gametangium of 8 cells in two rows: one nucleus in late prophase, polar view of which is shown in the previous figure; nucleus in basal cell in early prophase.

FIG. 75.—Portion of a female gametangium of 7 cells: one nucleus is in late prophase, showing 24 chromosomes, which will result in two cells arranged side by side in the direction perpendicular to the plane of the paper.

FIG. 76.—Portion of a female gametangium of 8 cells: one nucleus in anaphase.

FIG. 77.—Portion of a female gametangium of 8 cells: one nucleus in late anaphase.

FIG. 78.—Cross-section of a female gametangium of 10 cells: one nucleus in metaphase of the division which divides the gametangium into a structure of three rows of cells.

FIG. 79.—Portion of a female gametangium of 10 cells: one nucleus in metaphase.

FIGS. 80, 81.—Portion of a female gametangium of 10 cells: one nucleus in anaphase in both figures.

FIG. 82.—Portion of a female gametangium of 10 cells: one nucleus is in metaphase and two others have already divided.

FIG. 83.—Portion of a female gametangium of 10 cells: one nucleus in prophase, showing 24 chromosomes, one nucleus in late anaphase and the other two have already divided.

FIG. 84.—Portion of a female gametangium of 10 cells: one nucleus in early metaphase viewed from the pole, showing 24 chromosomes.

FIG. 85.—Portion of a female gametangium of 12 cells: one nucleus in anaphase and the other two have already divided.

FIG. 86.—Portion of a female gametangium of 12 cells: one nucleus in early metaphase, viewed from the pole, showing 24 chromosomes; the other nucleus in metaphase in lateral view.

FIG. 87.—Portion of a female gametangium of 15 cells: one nucleus in prophase.

FIG. 87*a*.—The nucleus in prophase in the previous figure under higher magnification: 24 chromosomes present.

FIGS. 88, 88*a*.—Portion of a female gametangium of 18 cells: one nucleus in anaphase, which is shown under higher magnification in fig. 88*a*.

FIG. 89.—Cross-section of mature female gametangium in 4 tiers.

FIG. 90.—Cross-section of mature female gametangium in 8 tiers.

FIG. 91.—Two cells of a mature female gametangium: the plastids and dense cytoplasm are thick along the inner wall; nuclei in resting condition.

FIG. 92.—One cell of a mature female gametangium: plastids and cytoplasm are thick toward the outer wall; nucleus in resting condition.

FIG. 93.—A female gametangium entirely emptied after the escape of female gametes.

PLATE XXIX

The union of gametes and germination of the fertilized female gamete

FIG. 94.—Two male gametes which have stopped the swimming movement; cilia are withdrawn, nuclear membranes are scarcely visible, and the reticulum shows 24 chromosomes.

FIG. 95.—Union of male and female gametes: nucleus of the male gamete shows 24 chromosomes and that of the female gamete is in the resting condition; no cell membrane is recognizable around the gametes.

FIG. 96.—Cytoplasm of male gamete is entirely fused with that of female gamete and the whole body of the united gametes has about assumed the spherical form; cell membrane has appeared; male nucleus still shows 24 chromosomes.

FIG. 97.—Male nucleus has advanced toward the female nucleus.

FIG. 98.—Male nucleus has moved nearer toward the female nucleus.

FIG. 99.—Male nucleus is attached to the female nucleus.

FIG. 100.—Male nucleus, showing 24 chromosomes, and female nucleus in resting condition.

FIG. 101.—Male nucleus very closely applied to the female nucleus; 24 chromosomes of the male nucleus still recognizable; a part of the cell wall of the sporeling has thickened.

FIG. 102.—Part of the male nucleus, evidently consisting exclusively of chromosomes, has become submerged in the female nucleus, which is in the resting condition.

FIG. 103.—Male nucleus has completely entered into the female nucleus; dense chromatin granules are to be seen at the part of female nucleus where the male nucleus has entered.

FIG. 104.—Fusion nucleus: network contains a number of chromatin granules that show no distinction between those derived from the male and female nuclei.

FIG. 105.—Fusion nucleus is in the resting condition.

FIG. 106.—Prophase: 48 chromosomes and a nucleolus are present; the chromosomes are apparently alike both in form and size; the sporeling shows an elongation at a point where the cell wall is thickened.

FIG. 107.—Metaphase: thickening of cell wall is present, though the elongation of the sporeling has not yet begun.

FIG. 108.—Fusion nucleus in very early prophase: characteristic elongation of the sporeling and thickening of the corresponding cell wall at that part is remarkable.

FIG. 109.—Portion of the elongated part of a sporeling with a resting nucleus.

FIG. 110.—Cross-section of the elongated portion of a sporeling at the same stage as the previous figure; fusion nucleus in prophase; 48 chromosomes are present.

FIG. 111, *a*, *b*.—Two sections of a sporeling: nucleus in prophase, showing 48 chromosomes; elongation and correponding thickening of the cell wall are evident.

FIG. 112.—Metaphase: Elongation of the sporeling and thickening of the cell wall at the point of the elongation are marked; axis of the figure is in the direction of the elongation.

FIG. 113.—Cross-section of a sporeling perpendicular to the axis of elongation; nucleus in late metaphase.

FIG. 114.—Cross-section of a sporeling at about same stage as in fig. 112; nucleus in metaphase is seen from the pole; 48 chromosomes are present.

FIG. 115.—Sporeling in 2-celled stage: two nuclei in resting condition.

FIG. 116.—Sporeling in 2-celled stage: nucleus in terminal cell in prophase, and that of basal cell in resting condition.

FIG. 117.—Sporeling in 2-celled stage: nucleus in terminal cell very small and in resting condition, while that of basal cell shows very early prophase.

FIG. 118.—Cross-section through a basal cell of a sporeling in 2-celled stage: nucleus is in late prophase; 48 chromosomes present.

FIG. 119.—Sporeling in 2-celled stage: nucleus in terminal cell in early prophase, and that of basal cell in resting condition.

FIG. 120.—Sporeling in 2-celled stage: nucleus in terminal cell in prophase, showing 48 chromosomes.

FIG. 121.—Sporeling in 3-celled stage: nucleus in basal cell in prophase, showing 48 chromosomes.

FIG. 122.—Sporeling in 3-celled stage: nucleus in basal cell in metaphase.

FIG. 123.—Sporeling in 3-celled stage: all the nuclei in prophase, showing 48 chromosomes.

PLATE XXX

The germination of the unfertilized female gamete

FIG. 124.—A female gamete in the resting condition: cell wall does not seem to be developed excepting at the point where a slight elongation is noticeable.

FIG. 125.—Cell wall is now recognizable, especially at the elongated point; nucleus in resting condition.

FIG. 126.—Nucleus with 24 chromosomes is prophase.

FIG. 127.—Nucleus in metaphase: contour of sporeling almost spherical.

FIG. 128.—Polar view of same stage as in previous figure: 24 chromosomes.

FIG. 129.—Polar view of metaphase showing 24 chromosomes: characteristic elongation and the thickened cell wall of the elongated part are recognizable.

FIG. 130.—Metaphase of same stage as in previous figure: axis of the figure is perpendicular to that of the previous one.

FIG. 131.—Anaphase.

FIG. 132.—Metaphase of the similar stage as in figure 130, but axis of the figure is perpendicular to that.

FIG. 133.—Anaphase.

FIG. 134.—Late telophase: elongation of the sporeling is remarkable.

FIG. 135.—Sporeling of 2-celled stage: nuclei in resting condition.

FIG. 136.—Sporeling of 2-celled stage: nucleus of terminal cell in prophase; 24 chromosomes present.

FIG. 137.—Sporeling of 2-celled stage: nucleus of terminal cell in metaphase viewed from pole; nucleus of basal cell in prophase; 24 chromosomes present.

FIG. 138.—Sporeling of 2-celled stage: nucleus of terminal cell in prophase; form of the chromosomes is rather long.

FIG. 139.—Sporeling of 2-celled stage: nucleus in basal cell in anaphase.

FIG. 140.—Sporeling of 2-celled stage: nucleus in terminal cell in late telophase and that of basal cell in metaphase, showing 24 chromosomes in polar view.

FIG. 141.—Sporeling in 3-celled stage: nucleus of basal cell in prophase, showing 24 chromosomes.

FIG. 142.—Sporeling in 3-celled stage: nucleus of middle cell in prophase, showing 24 chromosomes.

FIG. 143.—Sporeling in 3-celled stage: nucleus of basal cell in metaphase.

FIG. 144.—Sporeling of 12-celled stage.

FIG. 145.—Sporeling of 13-celled stage.

FIG. 146.—Sporeling of 22-celled stage.

PLATE XXXI

The formation of the zoosporangium of Aglaozonia reptans

FIG. 147.—Portion of a young thallus, showing a single layer of superficial cells and the larger cells below.

FIG. 148.—Portion of a young thallus, showing a single layer of superficial cells, hypodermal cells, and the larger cells below.

FIG. 149.—Portion of thallus in more advanced stage.

FIG. 150.—Portion of thallus in still more advanced stage: superficial cells have grown in size and will be zoosporangia or zoospore mother cells.

FIG. 151.—Young zoospore mother cell and stalk cell.

FIG. 152.—Zoospore mother cell of characteristic club-shape; nucleus in resting condition.

FIG. 153.—Resting nucleus slightly increased in size.

FIG. 154.—Resting nucleus with radiations at one pole.

FIG. 155.—Section of a mother cell cut obliquely: resting nucleus with radiations at two poles.

FIG. 156.—Nucleus in very early prophase, situated at one side, with a centrosome-like structure but no radiations.

FIG. 157.—Nucleus situated at the upper part of the mother cell with two centrosome-like structures and radiations.

FIG. 158.—Nucleus situated at the upper part with a centrosome-like structure and radiations.

FIG. 159.—Nucleus situated at the middle part with no radiations.

PLATE XXXII

The formation of the zoosporangium of Aglaozonia reptans (continued)

FIG. 160.—Nucleus of a zoospore mother cell in prophase, chromatin network irregularly traversing the nuclear cavity.

FIGS. 161–168.—Nucleus in synapsis.

FIG. 161.—Chromatin threads have begun to be arranged in form of loops which become attached by their ends to a part of the nuclear membrane.

FIG. 162.—Formation of loops is further advanced.

FIG. 163.—Cross-section of a mother cell; attachment of loops is at the side.

FIG. 164.—Attachment of loops to the nuclear membrane is all the way around.

FIG. 165.—Some of the loops are quite shortened and thickened, and yet some others are left behind and can be recognized running across the cavity.

FIG. 166.—Section near the base of crowded loops in contact with the membrane, showing 48 or more isolated cut sections of the loops.

FIG. 167.—Loops have shortened and thickened.

FIG. 168.—Nucleus emerging from synapsis: two pairs of chromatin loops are already in the form of chromosomes.

FIG. 169.—Majority of the two arms of each of these loops are going to form paired arms of bivalent chromosomes.

FIG. 170.—Diakinesis stage: 24 bivalent chromosomes are present; no polar radiations.

FIG. 171.—Spindle with two poles has just formed.

FIG. 172.—Metaphase.

FIG. 173.—Polar view of the metaphase, showing 24 bivalent chromosomes at the equatorial plate.

PLATE XXXIII

The formation of the zoosporangium of Aglaozonia reptans (continued)

As the stage advances, gradual increase of the thickening of the cell wall at the tip of the mother cell is noticeable.

FIG. 174.—Late metaphase.

FIG. 175.—Anaphase: 24 chromosomes at each pole.

FIG. 176.—Anaphase, side view.

FIG. 177.—Anaphase, further advanced.

FIG. 178.—Telophase.

FIG. 179.—Later telophase: two daughter nuclei in resting condition.

FIG. 180.—Two daughter nuclei increased in size: each nucleus has radiations at the poles.

FIG. 181.—Upper nucleus has grown considerably larger than the lower one.

FIG. 182.—Two nuclei are in contact.

FIG. 183.—Upper nucleus is in prophase; lower one still in resting condition.

FIG. 184.—Two nuclei simultaneously in metaphase.

FIG. 185.—Anaphase.

FIG. 186.—Late anaphase: a group of chromosomes at one pole of the upper nucleus is not indicated in this figure.

PLATE XXXIV

The formation of the zoosporangium of Aglaozonia reptans (continued)

FIG. 187.—Telophase of the second division in the mother cell.

FIG. 188.—Late telophase.

FIG. 189.—Metaphase of the third division.

FIG. 190.—Telophase of the same: 8 nuclei present.

FIGS. 191–193.—Zoospore mother cells drawn from unstained preparations.

FIG. 191.—Nucleus in prophase: numerous granules of various sizes are recognizable; globular structures of larger size are plastids.

FIG. 192.—Four-nucleate stage: numerous granules still visible; plastids have increased in number.

FIG. 193.—Eight-nucleate stage: plastids have much increased in number and the thickness of the cell wall at the top of the mother cell is noticeable.

FIG. 194.—Cross-section of a mother cell at about same stage as in fig. 190; plastids mostly arranged near the cell membrane.

FIG. 195.—Portion of a mother cell near maturity: thickness of the cell wall at the top of the mother cell has increased; plastids moving toward the nuclei and surrounding them.

FIG. 196.—Later stage: plastids are around the nuclei.

FIG. 197.—Portion of a mother cell: cytoplasm has become detached from the cell wall and cleavage furrows have appeared, so that general outlines of individual zoospores are established.

FIGS. 198, 199.—Portion of a mother cell containing zoospores.

FIG. 200.—Portion of cell wall at the tip of a mother cell after maturity: the cell wall in this part is considerably thickened and swollen.

FIG. 201.—Later stage: the cell wall at the tip of the mother cell has disintegrated into two lamellae.

FIG. 202.—Further advanced stage approaching the discharge of zoospores: disintegration has still progressed and broken the cell wall into three lamellae.

PLATE XXXV

The germination of the zoospore of Aglaozonia reptans

FIG. 203.—Zoospore 2 hours after becoming quiescent: cell wall has just developed; resting nucleus with a centrosome-like structure and radiations.

FIG. 204.—Zoospore 20 hours after becoming quiescent: nuclear condition is about similar to fig. 203.

FIG. 205.—Zoospore 24 hours after becoming quiescent: no radiations visible.

FIGS. 206–208.—Drawn from material fixed 24 hours after becoming quiescent.

FIG. 206.—Resting nucleus with radiations: elongation of the sporeling and thickening of cell wall at the elongated part is noticeable.

FIG. 207.—About the same stage as in the previous figure.

FIG. 208.—Early prophase.

FIG. 209.—Prophase: 24 chromosomes clearly present.

FIG. 210.—Cross-section through the elongated part of a sporeling: nucleus in prophase, showing 24 chromosomes and a nucleolus.

FIG. 211.—Metaphase: general contour of the sporeling is spherical.

FIG. 212.—Polar view of the metaphase; 24 chromosomes clearly in view.

FIG. 213.—Metaphase: the sporeling has elongated.

FIG. 214.—Metaphase viewed from the pole: the sporeling has elongated at two points.

FIG. 215.—Anaphase.

FIG. 216.—Telophase.

FIG. 217.—Sporeling of 2 cells: nucleus of the basal cell is in prophase; 24 chromosomes visible.

FIG. 218.—Sporeling of 2 cells: nucleus in metaphase.

FIG. 219.—Sporeling of 2 cells: nucleus of terminal cell in early prophase; 24 chromosomes present.

FIG. 220.—Nucleus of terminal cell in prophase.

FIG. 221.—Nucleus of terminal cell in metaphase viewed from pole.

FIG. 222.—Two nuclei simultaneously in prophase; 24 chromosomes present.

FIG. 223.—Sporeling of 3 cells.

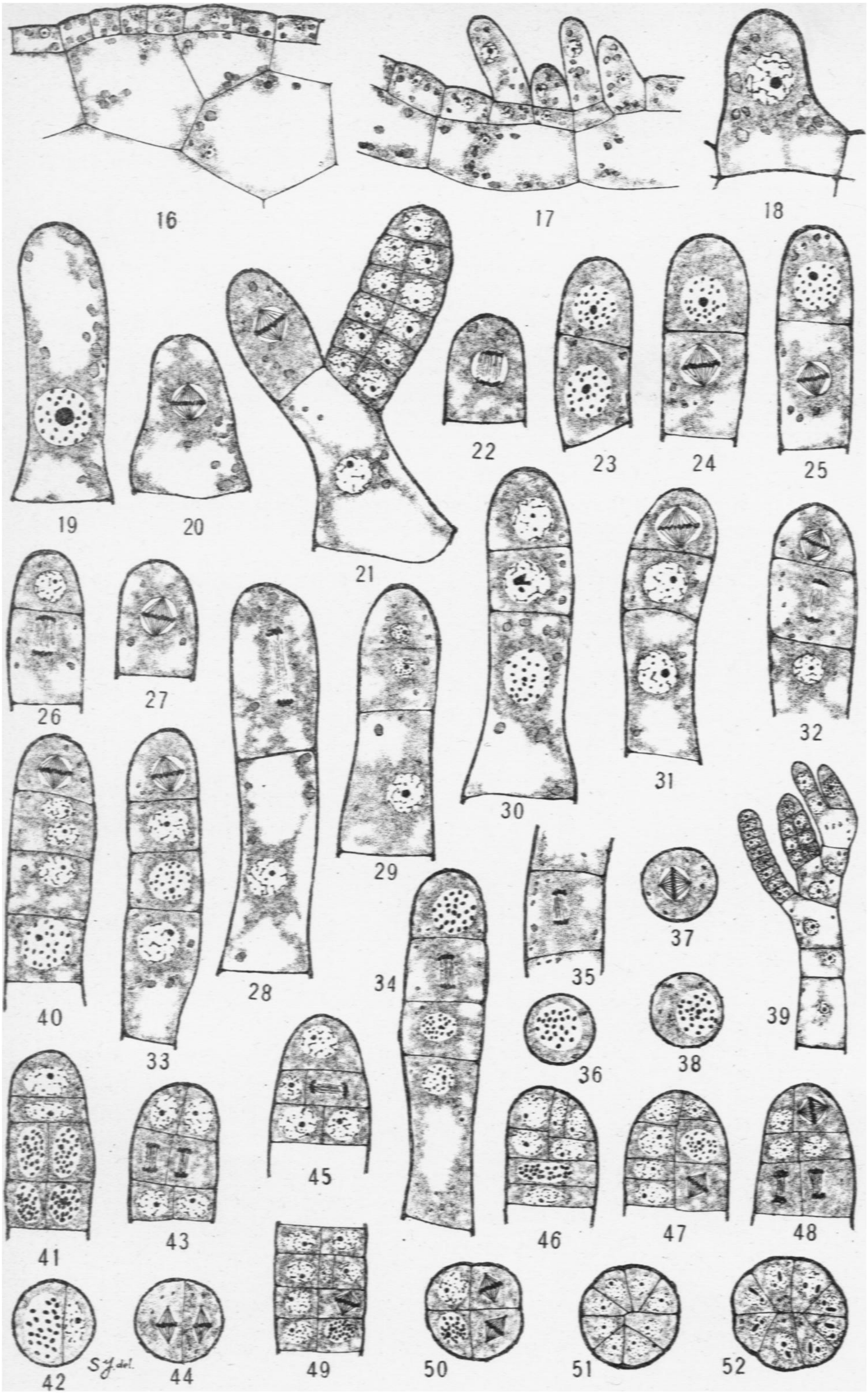
FIG. 224.—Sporeling of 3 cells: nucleus in middle cell in prophase; 24 chromosomes present.

FIG. 225.—Sporeling of 3 cells: nucleus in basal cell in prophase; 24 chromosomes present.

FIG. 226.—Sporeling of 4 cells.

FIG. 227.—Sporeling of 4 cells: nuclei in lower cells in metaphase.

FIG. 228.—Sporeling of 4 cells: two upper cells lie side by side in the direction perpendicular to the axis of the sporeling.



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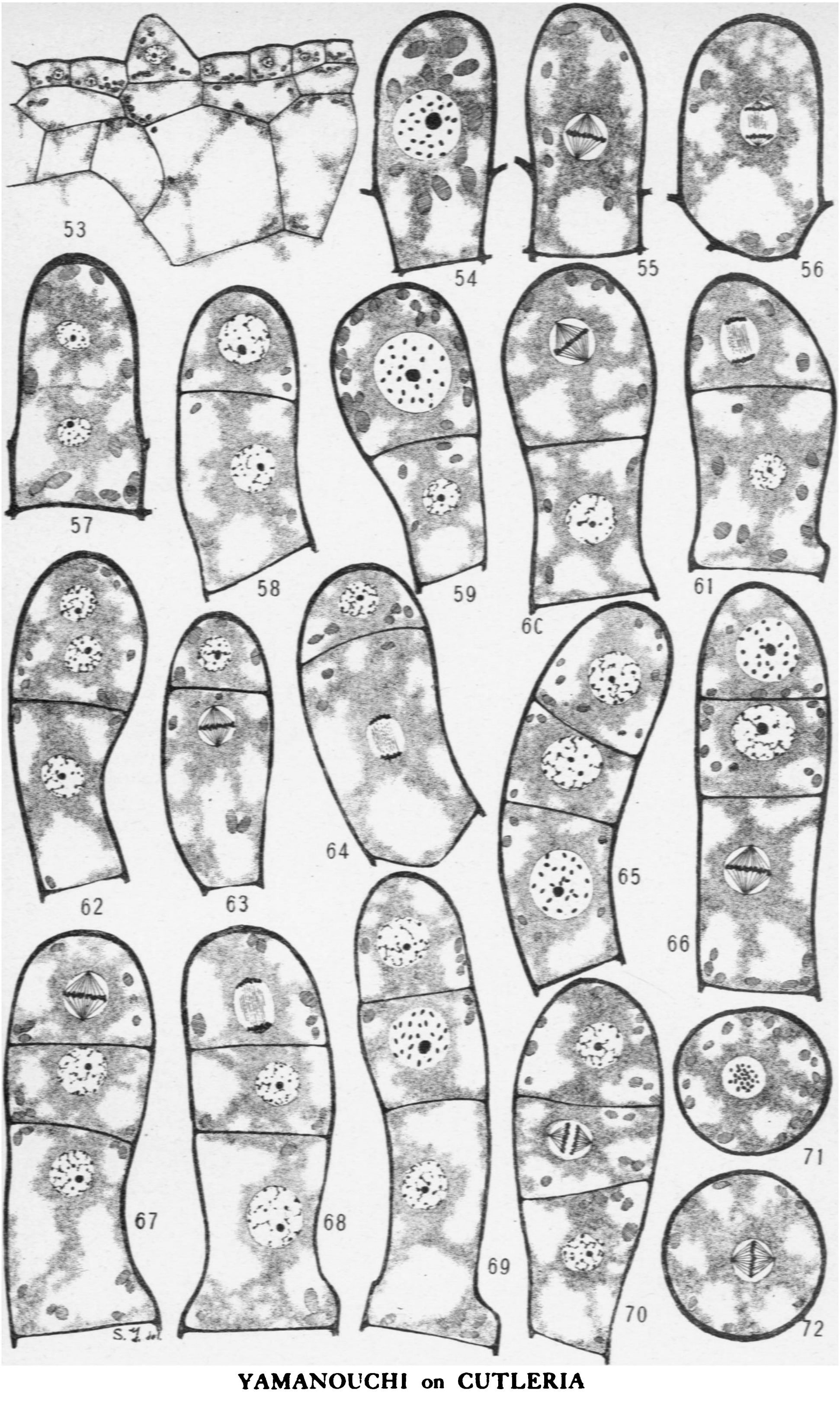

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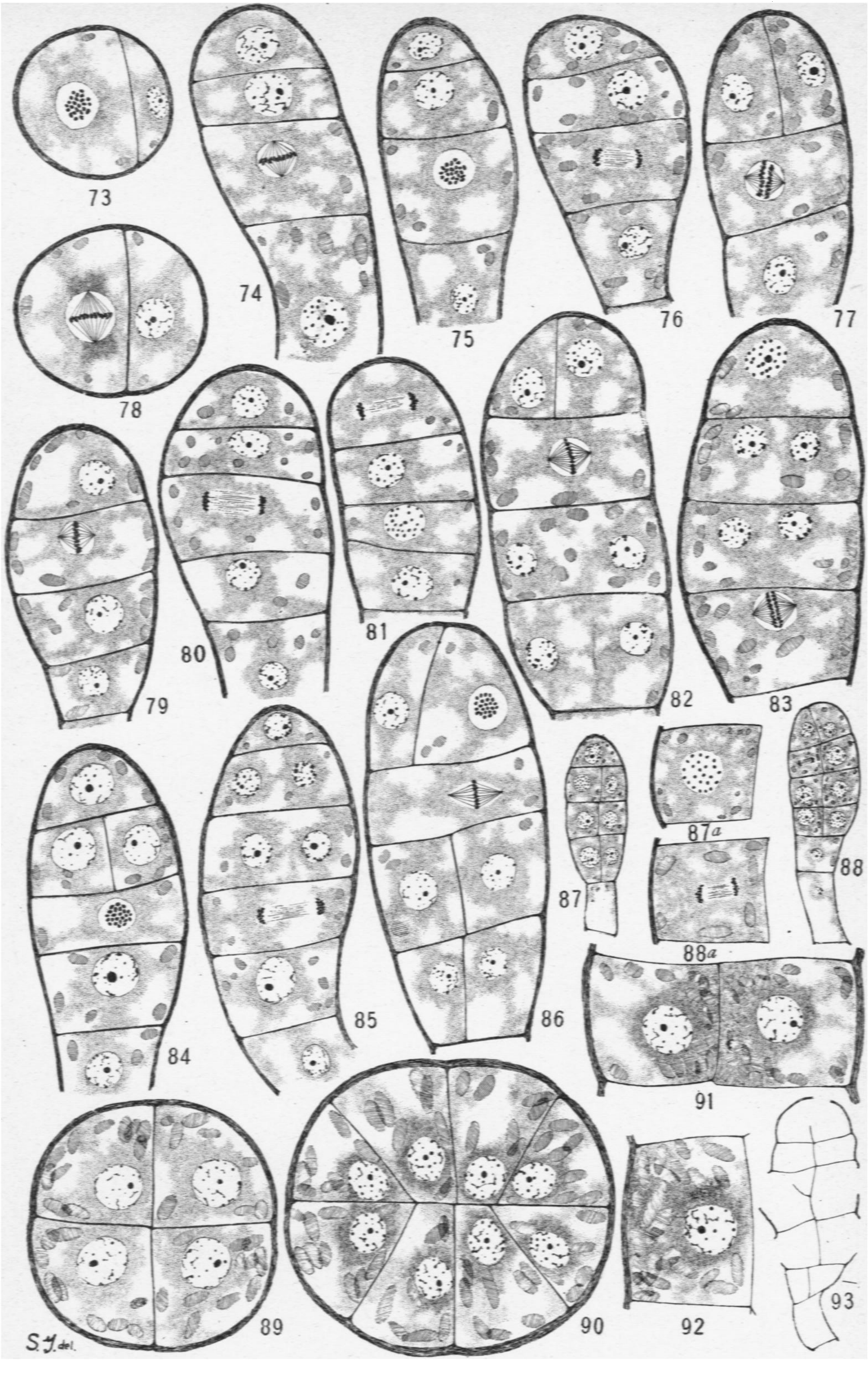

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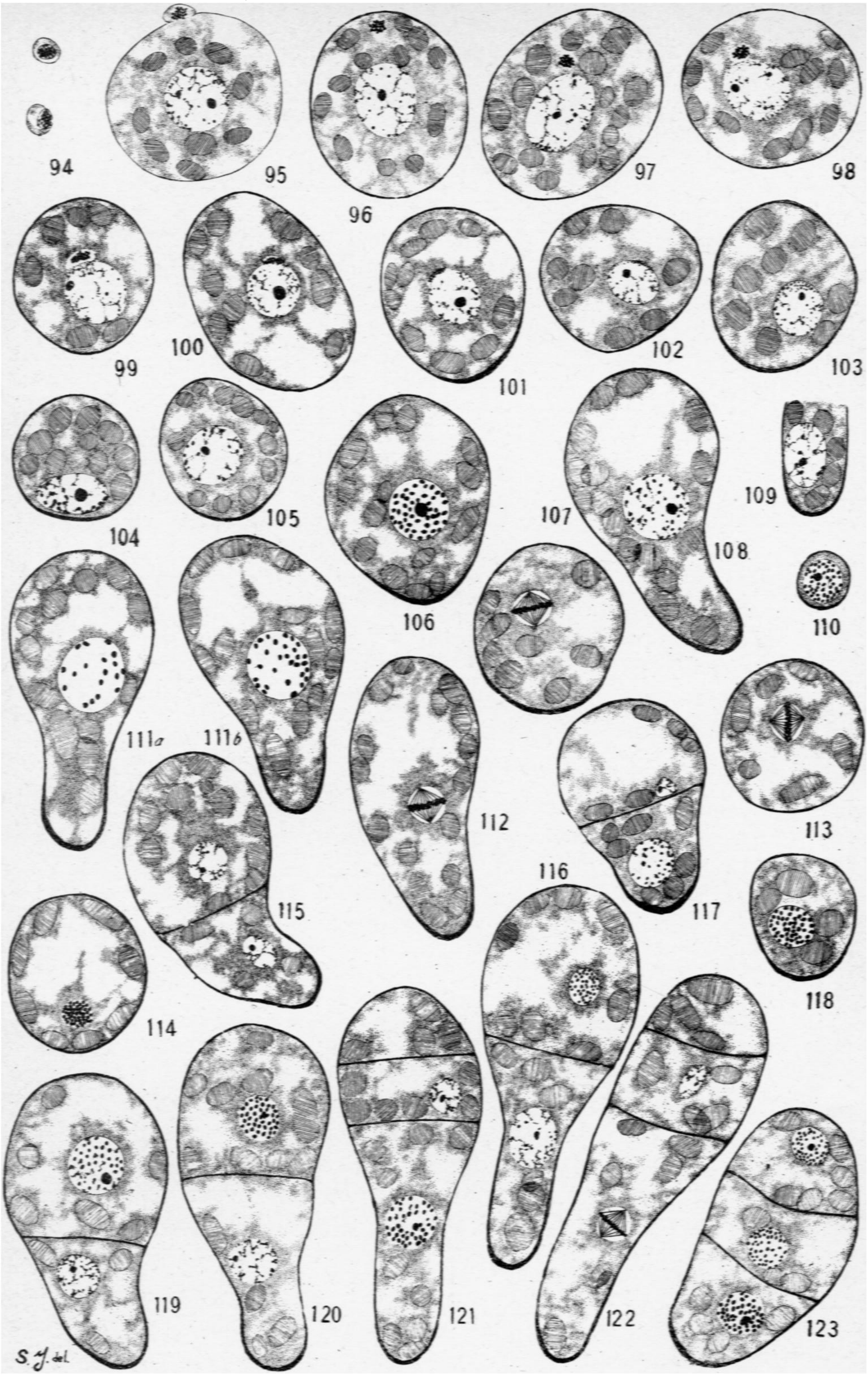

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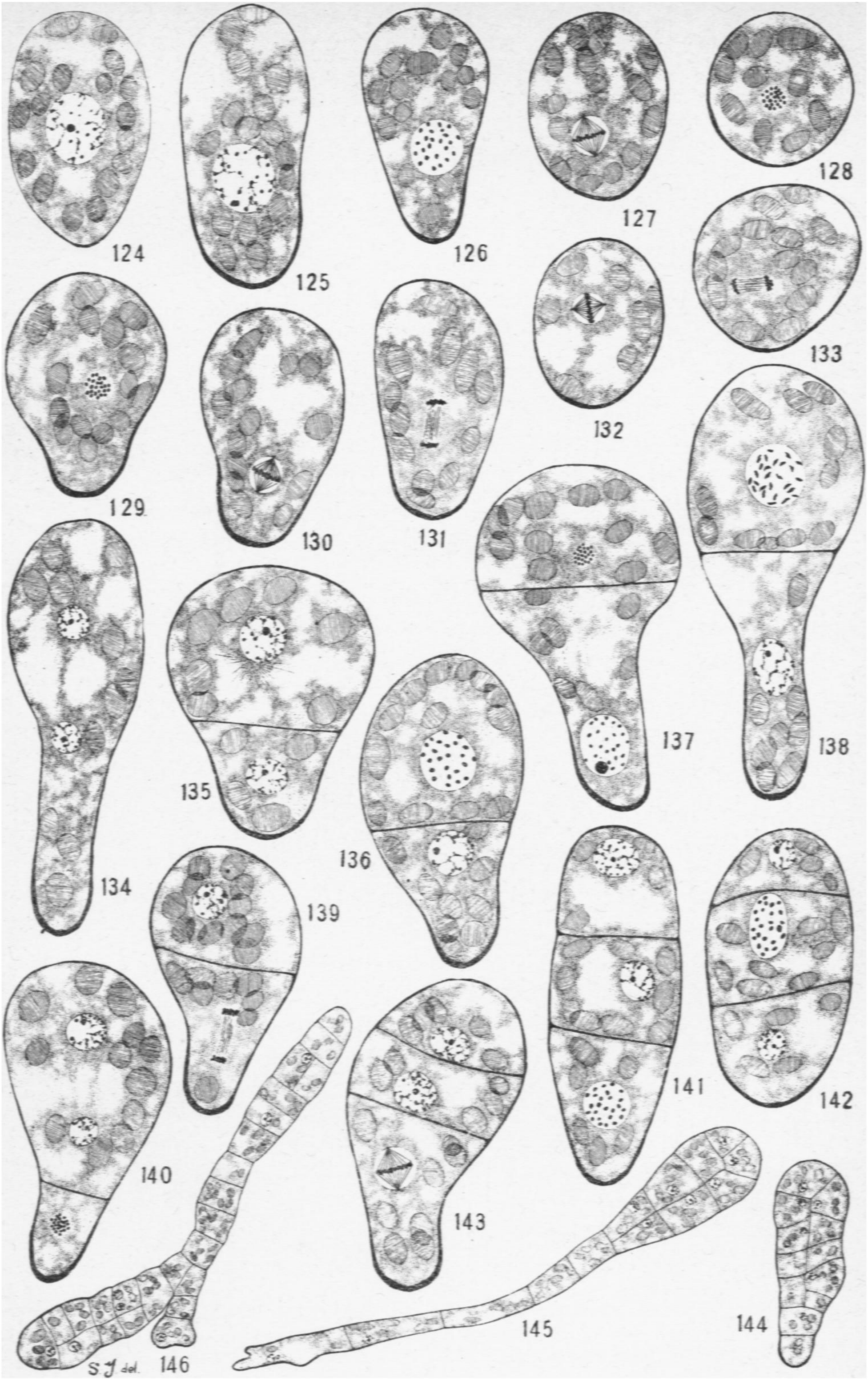

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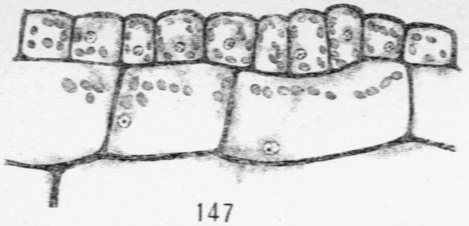
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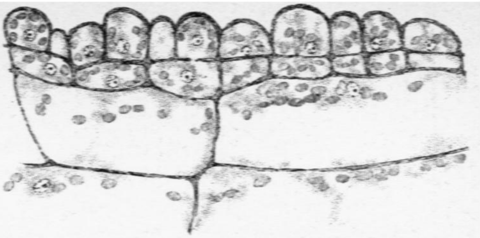
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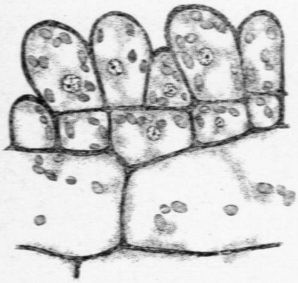
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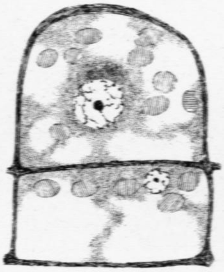
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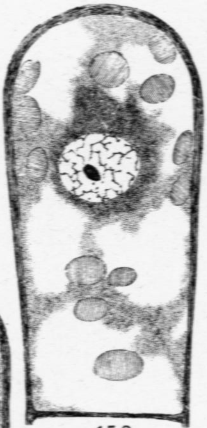
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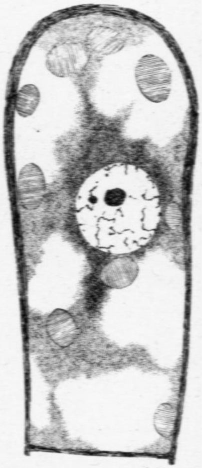
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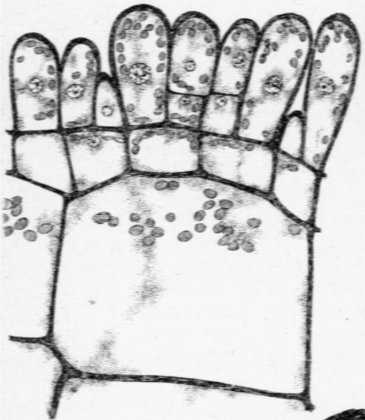
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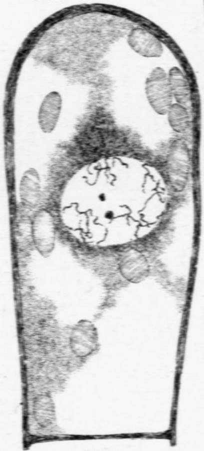
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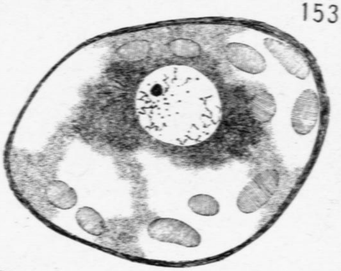
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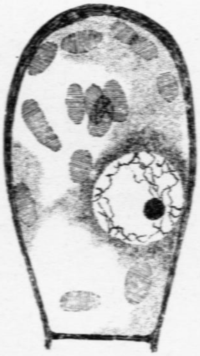
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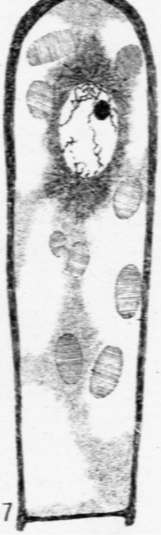
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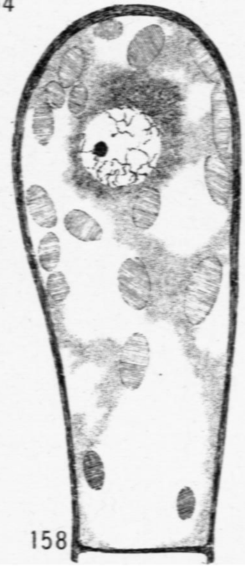
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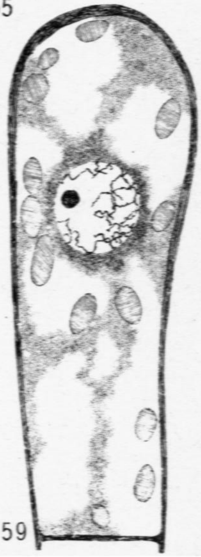
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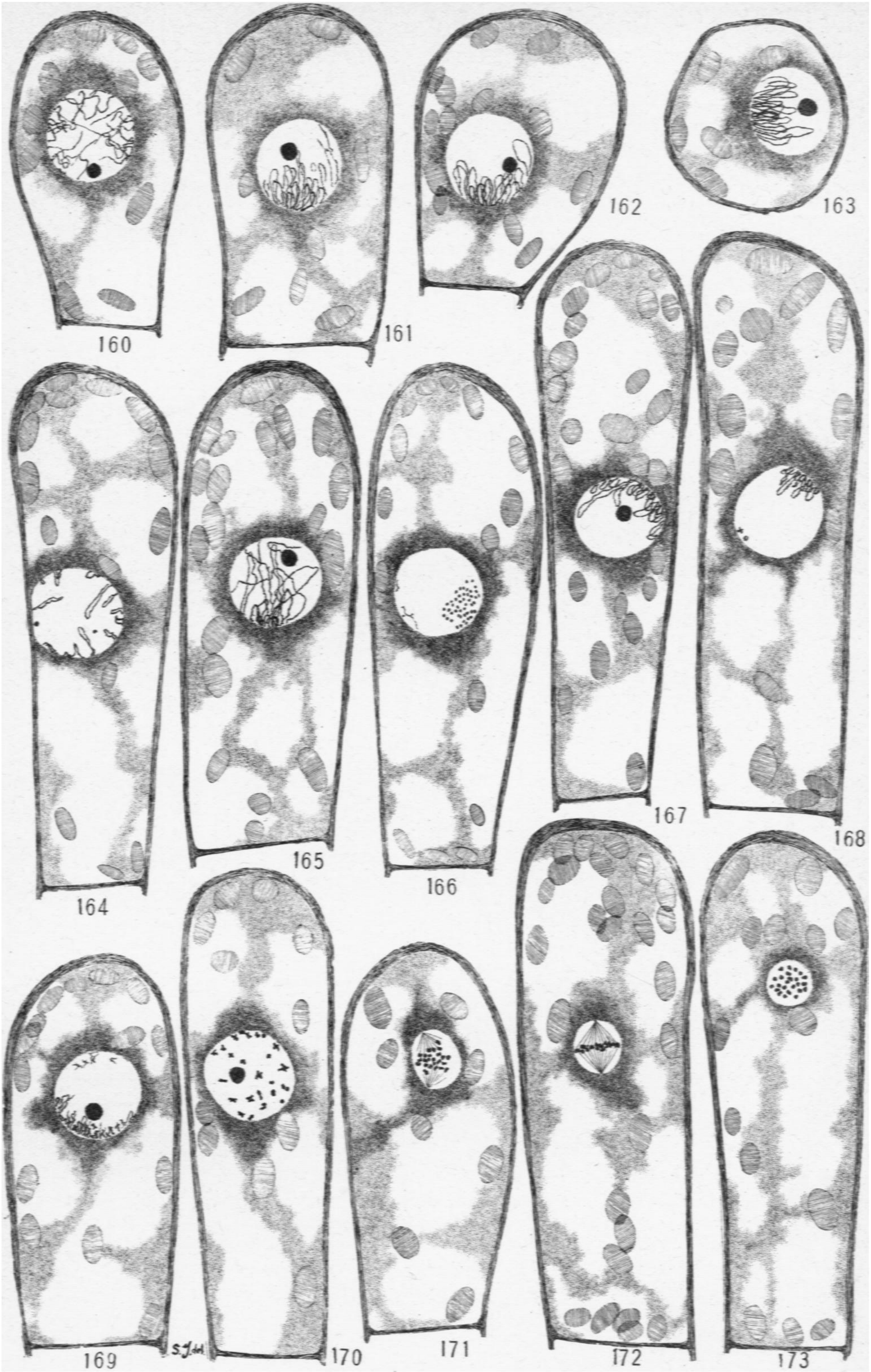

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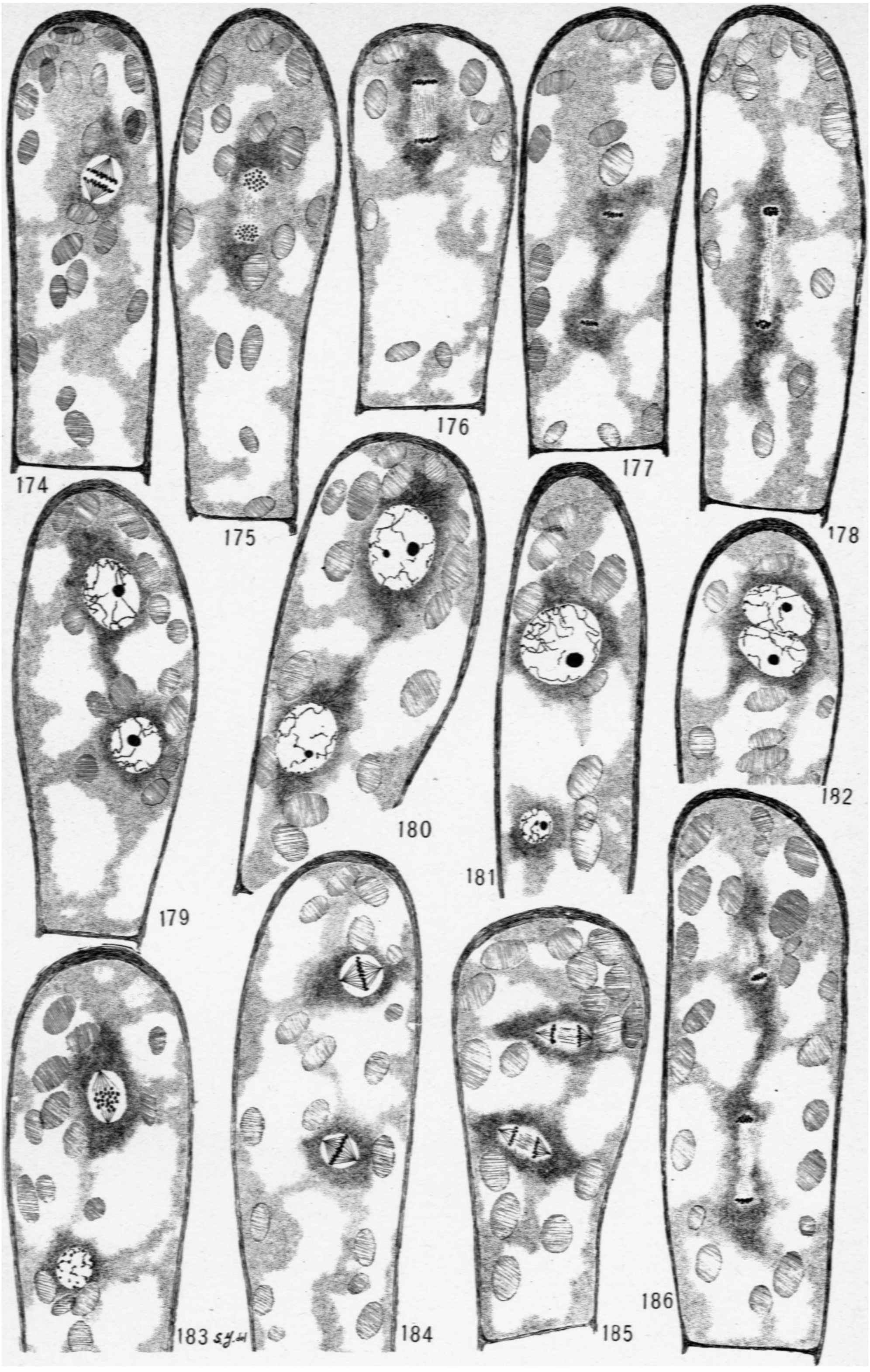

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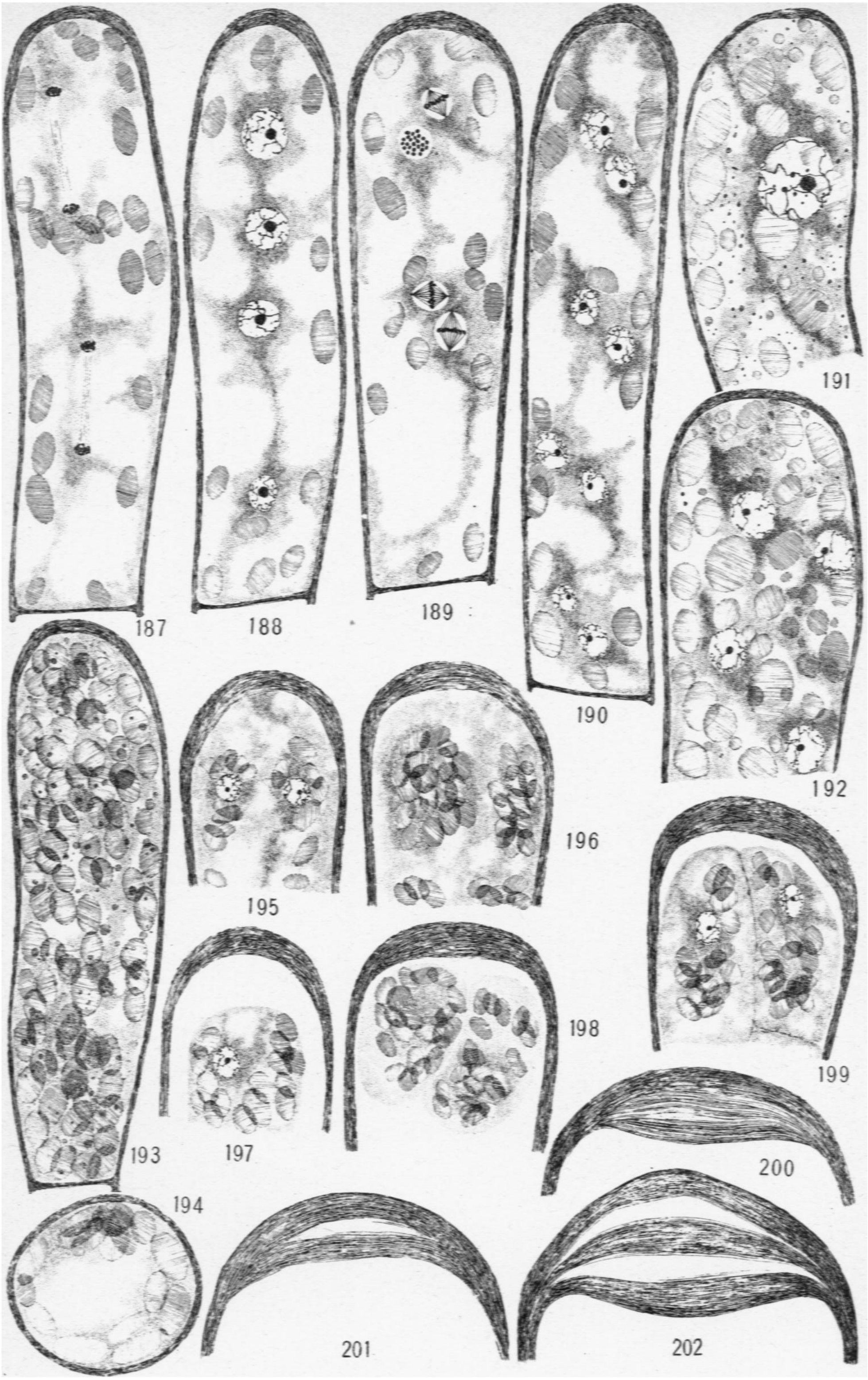

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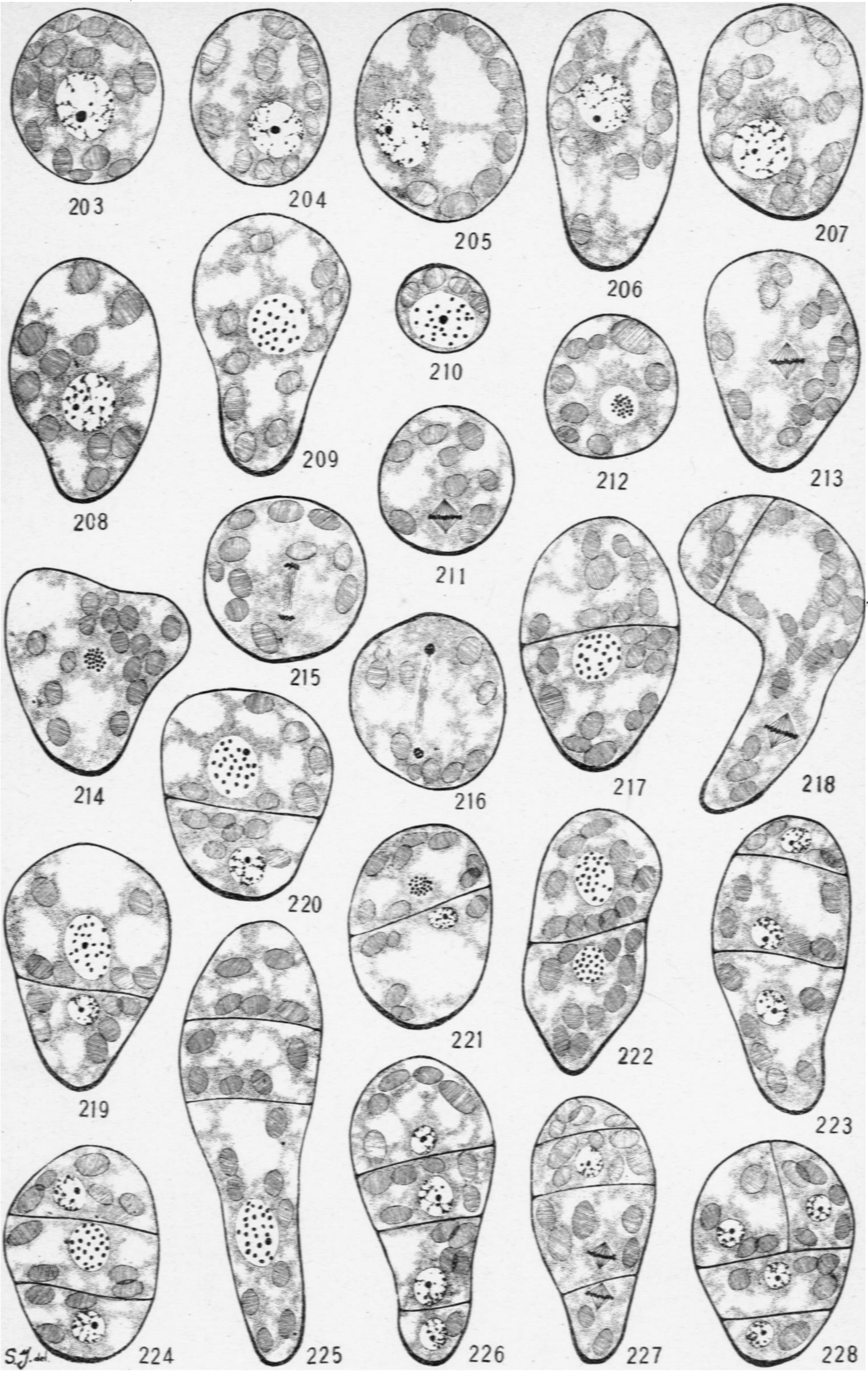

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